

H A N D B O O K O F

Microbiological
Media for the
Examination of Food

S E C O N D E D I T I O N

By

RONALD M. ATLAS



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Preface

Increased emphasis on the microbiological safety of foods has led to greater testing of foods for potentially pathogenic microorganisms. Careful quality control in the food industry has become essential for preventing outbreaks of foodborne disease and for controlling food spoilage. Many foods are routinely examined for the presence of disease-causing and food spoilage microorganisms. Many of these quality control procedures involve the cultivation of microorganisms using a variety of media that are described in the second edition of the *Handbook of Microbiological Media for the Examination of Food*.

Given that an estimated 14 million cases of foodborne disease with known microbial pathogens occur every year in the United States alone, the Food and Drug Administration and U.S. Department of Agriculture have been implementing new regulations and guidance for the microbial testing of a variety of foods. Similarly, Europe and other regions of the developed world are implementing stricter oversight of food safety to ensure that foods are free of potentially dangerous pathogens, e.g., *Salmonella* spp., *Vibrio* spp., *Listeria monocytogenes*, *Campylobacter jejuni*, etc.

Emerging diseases caused by foodborne pathogens, such as *Escherichia coli* O157:H7, that have high mortality rates have raised special concerns. This has encouraged the development of new media for the cultivation of bacteria, fungi, and viruses that can aid in the rapid detection of microorganisms in foods and the protection of the food supply. Many of these new media include chromogenic or fluorogenic substrates that permit the specific rapid detection of pathogens of concern.

The second edition of the *Handbook of Microbiological Media for the Examination of Food* provides descriptions of the media used to cultivate microorganisms from foods. Almost 1,400 media are described in the *Handbook*, including all the media recommended by the Food and Drug Administration for the detection of microorganisms in foods as well as media used elsewhere in the world. The *Handbook* includes descriptions of the media used to cultivate microorganisms used for food production as well as those that are important for quality control, i.e., the cultivation of potential pathogens and food-spoilage microorganisms.

The second edition of the *Handbook of Microbiological Media for the Examination of Food* compiles in one place in a consistent style the formulations,

methods of preparation, and uses of each medium. It lists the common synonyms for the various media and the commercial sources when they are available. The index provides a convenient tool for finding the media by names and application for the detection of specific microorganisms of interest.

Importantly, the *Handbook of Microbiological Media for the Examination of Food* includes descriptions of expected results as they apply to microorganisms of importance for the examination of foods. The compositions of various media can be compared so that alternate media can be used. Similar or identical media described by different names that actually have the same formulations can be readily identified.

Using the second edition of the *Handbook of Microbiological Media for the Examination of Food* should save time and effort for anyone cultivating microorganisms. The media are organized alphabetically. Each medium includes the composition, instructions for the preparation, and uses of each of the media.

The information needed to prepare media for the cultivation of microorganisms can easily be found in the *Handbook*. It should be a valuable resource for anyone working in the area of food microbiology.

About the Author

Ronald M. Atlas is Professor of Biology at the University of Louisville. He received his B.S. from the State University of New York at Stony Brook in 1968 and his M.S. and Ph.D. from Rutgers in 1970 and 1972, respectively. Dr. Atlas has received a number of honors including: The University of Louisville Excellence in Research Award, the Johnson and Johnson Fellowship for Biology, and the ASM Award in Applied and Environmental Microbiology.

He has taught a variety of courses in microbiology including food and industrial microbiology and has authored several microbiology textbooks. He has conducted studies on the use of gene probes and the polymerase chain reaction for the detection of pathogens and indicator bacteria in water and food. He has served on the editorial boards of *Applied and Environmental Microbiology*, *BioScience*, *Biotechniques*, and *Journal of Industrial Microbiology*. He is editor of *CRC Critical Reviews in Microbiology*. He also has served as President of the American Society for Microbiology.

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Introduction to Food Microbiology

Food products serve not only as sources of nutrition for humans and other animals but also as substrates for the growth of microorganisms. The uncontrolled growth of microorganisms in food causes spoilage, a serious problem accounting for sizable losses of food products. Some disease-causing microorganisms also contaminate foods and potable waters. Foodborne and waterborne pathogens remain a major source for the spread of disease worldwide. A major concern in food microbiology is the preservation of food products and the prevention of foodborne and waterborne spread of disease. Careful quality control in the food industry is essential for preventing outbreaks of foodborne disease and for controlling food spoilage. Many foods are routinely examined for the presence of disease-causing and food spoilage microorganisms. Many of these quality control procedures involve the cultivation of microorganisms using a variety of media.

Food- and Waterborne Pathogens

The ingestion of foods containing toxins and human pathogens can cause a variety of diseases. It is essential to prevent contamination of food products with human pathogens and to control the potential proliferation of toxin-producing microorganisms, which can result in food poisoning and the transmission of foodborne pathogens. In some cases, the growth of pathogenic microorganisms in a food is accompanied by obvious signs of spoilage, such as the production of gas or foul-smelling compounds, which give a clear indication that the product should not be eaten. In other cases, there are no obvious signs of spoilage to indicate the presence of pathogens or toxins. It is therefore necessary to carry out inspection programs aimed at detecting the possible contamination of food products with pathogenic microorganisms. As a consequence of the high quality control procedures employed in the food industry, few occurrences of foodborne disease are traced back to the food processor. Rather, most outbreaks of food poisoning are caused by improper postprocess handling of the food, often as a result of improper preparation and/or handling at home or in a food service setting.

Quality control laboratories in the food industry routinely perform tests to detect pathogens in food products and to ensure that the numbers and types of microorganisms associated with a food are not likely to cause serious food spoilage and/or health problems. In some cases, quality control procedures are aimed at detecting the presence of specific microorganisms, but in most cases the tests consist of exam-

ining the food for indicator organisms. For example, coliform counts are routinely performed on representative samples of many food products as an indication of possible fecal contamination, since food contaminated with fecal material has a relatively high probability of containing human pathogens. Foods such as hamburger often have 100,000 total bacteria per gram, but as long as they do not contain any *Salmonella* or other pathogens, they are considered safe for human consumption.

In the United States, agencies involved in the surveillance and regulation of foods are the Department of Agriculture (USDA), the Food and Drug Administration (FDA), and state boards of health. Standards based on the presence or absence of pathogens, such as those set by Congress and regulated by the USDA and FDA, do provide the safeguards needed for assurance of the safety of food products. On the international front, standards for the microbial testing of foods are established by the International Organization for Standardization (ISO). Both the FDA and ISO standards specify the media and procedures that are to be employed for the microbial analyses of specific foods.

Microorganisms routinely enter the gastrointestinal tract in association with ingested food and water. The large resident microbiota that develops in the human intestinal tract after birth is important for the maintenance of good health and is usually not involved in disease processes. This microbiota is normally noninvasive and is associated with the surface tissues and ingested food material. Some pathogenic microorganisms, however, possess toxigenic or invasive properties that permit them to cause disease when they enter the gastrointestinal tract.

There are two distinct processes that can initiate disease through the gastrointestinal tract. In the first type, microorganisms growing in food or water can produce toxins, and their ingestion initiates a disease process. Such diseases are classified as food poisoning or intoxication because the etiological agents of the disease need not grow within the body; that is, there is no true infectious process. Toxins absorbed through the gastrointestinal tract can cause neural damage and death in some cases, as well as localized inflammation and gastrointestinal upset in others. In the second type of disease-causing process, invasive pathogens establish an initial infection through the gastrointestinal tract and cause localized gastrointestinal upset or systemic disease symptomatology. Generally, the establishment of infection through the gastrointestinal tract requires a relatively large infec-

tious dose; that is, a relatively large number of pathogenic microorganisms is required to successfully overcome the inherent defense mechanisms of the gastrointestinal tract. Quite different measures are required to prevent and treat infectious gastrointestinal diseases compared to those for specific microorganisms responsible for food poisoning.

Many outbreaks of gastroenteritis involve the foodborne or waterborne transmission of viruses that can replicate within the human gastrointestinal tract and cause an inflammation of the lining of the gastrointestinal tract. Contamination of food with fecal matter is an important route of transmission of viral gastroenteritis. Several different viruses cause viral gastroenteritis, including adenoviruses, coxsackieviruses, polioviruses, and members of the ECHO virus group. The Norwalk agent, a small DNA virus identified as being responsible for an outbreak of "winter vomiting disease" that occurred in Norwalk, Ohio, in 1968, appears to be an important etiological agent of various viral gastroenteritis outbreaks. Rotavirus, a large RNA virus, also appears to be a very common etiological agent of diarrhea in infants, particularly in socioeconomically depressed regions of the world. It is difficult to detect the diversity of viruses causing this disease and foods and waters are only rarely examined for their presence.

Hepatitis is a systemic viral infection that primarily affects the liver, caused by several types of hepatitis viruses, designated types A, B, C, D, and non-A, non-B. Type A hepatitis virus is normally associated with foodborne and waterborne cases of hepatitis. Type A hepatitis virus normally enters the body via the gastrointestinal tract and causes infectious hepatitis. Although there are some documented cases of foodborne hepatitis B, types B, D and non-A, non-B hepatitis viruses, these viruses normally enter the body through skin punctures and cause serum hepatitis. Hepatitis type A virus is usually transmitted by the fecal-oral route and is prevalent in areas with inadequate sewage treatment. Several outbreaks of viral hepatitis have been associated with contaminated shellfish, such as oysters, that have concentrated viruses from sewage effluents. There have also been outbreaks from vegetables contaminated with infected fecal matter used as fertilizer. Contaminated potable water supplies are probably the most common source of hepatitis A viral infections.

Polioviruses may enter the body through either the gastrointestinal or respiratory tracts. Transmission through the ingestion of food and water containing polioviruses is considered very important. Polio is effectively controlled through the use of vaccines. This has been responsible for the decline of this dis-

ease, particularly in developed nations. Global vaccination programs are aimed at eliminating this disease worldwide.

Several *Clostridium* species cause foodborne disease. Canned foods are often the sources of clostridia causing human disease because *Clostridium* species are endospore forming anaerobes. Hence they are resistant to elevated temperatures, sometimes surviving the canning process high temperature killing, and can grow in the can because of the absence of air. Botulism, the most serious form of bacterial food poisoning, is caused by neurotoxins produced by *Clostridium botulinum*. The toxins are absorbed from the intestinal tract and transported via the circulatory system to motor nerve synapses, where their action blocks normal neural transmissions. Various strains of *C. botulinum* elaborate different toxins. Types A, B, and E toxins cause food poisoning of humans. Type E toxins are associated with the growth of *C. botulinum* in fish or fish products, and most outbreaks of botulism in Japan are caused by type E toxins because large amounts of fish are consumed there. Type A is the predominant toxin in cases of botulism in the United States, and type B toxin is most prevalent in Europe. *C. botulinum* cultures form three distinct physiological groups that can be detected by culture methods. *C. botulinum* types C and D are nonproteolytic and do not digest coagulated egg white or meat. All type A and some type B and F strains of *C. botulinum* are proteolytic whereas all type E strains and the remaining type B strains are nonproteolytic. Type G strains of *C. botulinum* show slow proteolysis. The optimum temperature for toxin production by proteolytic strains is approximately 35°C and for nonproteolytic strains is about 27°C. Nonproteolytic strains of *C. botulinum* types B, E, and F produce toxin at 4°C.

Over 90% of the cases of botulism involve improperly home-canned food. Of 236 outbreaks of this disease in the United States between 1899 and 1974, 57% were caused by contaminated vegetables, 15% by contaminated fish, and 12% by contaminated fruit. Certain canned foods provide an optimal anaerobic environment for the growth of *C. botulinum* that results in the release of toxin into the food. *C. botulinum*, though, cannot grow and produce toxin at low pH and thus is not a problem in acidic food products. Another clostridial species, *Clostridium perfringens*, is a major cause of a less severe form of food poisoning. *C. perfringens* generally accounts for over 10% of the outbreaks of foodborne disease in the United States. The ingestion of food containing toxin produced by *C. perfringens* and the adsorption of the toxin into the cells lining the gastrointestinal tract ini-

tiate this disease. Toxin type A of *C. perfringens* is associated with most cases of clostridial food poisoning, particularly with cooked meats if a gravy is prepared with the meat. The spores of *C. perfringens* type A can survive the temperatures used in cooking many meats, and if incubated in a warm gravy, there is sufficient time for the spores to germinate and the growing bacteria to produce enough toxin to cause this disease.

Bacillus cereus is responsible for a relatively mild form of gastroenteritis, and recovery normally occurs in less than a day. The occurrence of gastroenteritis due to *B. cereus* requires the ingestion of a large number of spores. The symptoms include abdominal pain, profuse diarrhea, and nausea. *B. cereus* foodborne disease most commonly is associated with custards, cereal products, and meat gravies.

Strains of *Staphylococcus aureus* produce a toxin that causes an inflammation of the lining of the gastrointestinal tract. *S. aureus* can reproduce within many different types of food products. Enterotoxin-producing strains of *S. aureus* often enter foods from the skin surfaces of people who handle food. Custard-filled bakery goods, dairy products, processed meats, potato salad, and various canned foods are frequently found to be the source of the toxin. The prevention of staphylococcal food poisoning depends on proper handling and preservation of food products to prevent contamination and subsequent growth of enterotoxin-producing strains of *Staphylococcus*.

Listeria monocytogenes infects cattle, sheep, and chickens. It is transmitted to humans via contaminated milk and poultry and fecally-contaminated water and vegetables. Unpasteurized milk often is the source of human infection and cheeses have been a major source of *Listeria* infections in the United States. Identification of *Listeria monocytogenes* is based upon observation of Gram-positive rods that exhibit tumbling motility, utilization of lactose, catalase positive and oxidase negative reactions, exculin and sodium hippurate hydrolysis, Methyl Red positive reaction, ammonia production from arginine, and negative reactions for hydrogen sulfide production, nitrate reduction, gelatin liquefaction, starch hydrolysis, and urea hydrolysis.

Various *Salmonella* species, including *S. choleraesuis* and especially the numerous serotypes of *S. enteritidis*, are commonly the etiological agents of salmonellosis. Like many enteropathogenic bacteria, *Salmonella* species have pili that enable them to adhere to the lining of the gastrointestinal tract. Although *Salmonella* species are able to reproduce within the intestines, causing inflammation, they do not normally penetrate the mucosal lining and enter

the bloodstream; in some cases, however, *Salmonella* species can gain access to the circulatory system, causing bacteremia. For example, paratyphoid fever, which is caused by strains of *S. paratyphi* and *S. typhimurium*, is characterized by gastroenteritis and a relatively high rate of bacteremia. *Salmonella* species causing gastroenteritis are normally transmitted by ingestion of contaminated food. Birds and domestic fowl, especially ducks, turkeys, and chickens, including their eggs, are commonly identified as the sources of *Salmonella* infections. Inadequate cooking of large turkeys and the ingestion of raw eggs cause a significant number of cases of salmonellosis. Outbreaks of typhoid fever, a systemic infection caused by *Salmonella typhi*, are associated with contaminated water supplies and the handling of food products by individuals infected with this bacterium. A relatively low infectious dose is required for *S. typhi* to establish an infection. Some individuals develop a carrier state; such individuals do not develop typhoid fever but act as a source of *S. typhi* and spread the disease as in the infamous case of a cook in the early twentieth century who became known as typhoid Mary.

Shigellosis, or bacterial dysentery, is an acute inflammation of the intestinal tract caused by species of the Gram-negative genus *Shigella*, including *S. flexneri*, *S. sonnei*, and *S. dysenteriae*. Water and food supplies are involved in some outbreaks of bacterial dysentery. The severe dehydration associated with this disease can cause shock and lead to death in children, in whom the incidence of bacterial dysentery is highest.

Campylobacter fetus var. *jejuni* has been found to be the causative agent of many cases of gastroenteritis in infants. In fact, *C. fetus* may be more important in juvenile gastroenteritis than *Salmonella* species. The transmission of *C. fetus* appears to be via contaminated food or water. *C. fetus* is a Gram-negative, motile, spiral-shaped bacterium, formerly known as *Vibrio fetus*, which also causes fetal abortion in cattle and sheep.

Several *Vibrio* species, including *V. cholerae*, *V. parahaemolyticus*, *V. mimicus*, and *V. vulnificus* cause foodborne and waterborne diseases. *Vibrio* species are Gram negative straight or curved rods (single curve) and ferment glucose without producing gas. *V. vulnificus* is most often associated with seafoods as a source of foodborne infections. *V. parahaemolyticus* is responsible for many cases of gastroenteritis in Japan and perhaps in the United States. It occurs in marine environments, and the ingestion of contaminated seafood, particularly the eating of raw fish, is the main route of transmission. Gastroenteritis caused by *V.*

parahaemolyticus requires the establishment of an infection within the gastrointestinal tract, rather than simple ingestion of an enterotoxin. The symptoms generally appear 12 hours after ingestion of contaminated food and include abdominal pain, diarrhea, nausea, and vomiting. Recovery from this form of gastroenteritis normally occurs in 2 to 5 days, and the mortality rate is very low.

In contrast, cholera, which is caused by *Vibrio cholerae*, serotypes *cholerae* and *El Tor*, often is fatal. Although we typically associate cholera with Asia, sometimes referring to the disease as *Asiatic cholera*, it also occurs in the United States, primarily in the Gulf Coast region, where cases have been traced to contaminated shellfish. Cholera is now endemic to South America where eating of contaminated raw fish is a major source of infection. Contaminated water supplies are also responsible for the geographic spread of cholera from the initial focus in Peru throughout Latin America. Cholera is a particular problem in socioeconomically depressed countries, where there is poor sanitation and inadequate sewage treatment and where medical facilities have only a limited capacity to deal with outbreaks. This disease is endemic in the Ganges delta, and there are annual epidemic outbreaks of cholera in India and Bangladesh. In these endemic areas of Asia the death rate is normally 5 to 15%. Seasonal outbreaks of cholera often occur in Southeast Asia when monsoon rains wash sewage material into drinking water supplies. During sudden epidemics, the mortality rate may reach 75%.

Yersinia enterocolitica and related species, such as *Y. pseudotuberculosis*, produce a severe form of enterocolitis. Outbreaks of yersiniosis are most common in Western Europe but have also been confirmed in the United States. *Y. pseudotuberculosis* is found in pork, raw milk, and various other foods. *Y. enterocolitica* is widely distributed and has been found in water, milk, fruits, vegetables, and seafoods. This organism is psychrotrophic and thus is able to reproduce within refrigerated foods, where it can multiply and reach an infectious dose. In fact, *Y. enterocolitica* grows better at 25°C than at 37°C.

Coliform bacteria, principally *Escherichia coli*, are widely used as indicators of human fecal contamination of food and water. When used in this manner *E. coli* is not screened as a pathogen but rather because it is found in high numbers in human feces and is relatively short lived in the environment. Finding *E. coli* in food or potable water indicates contamination with human fecal matter that may also carry pathogens such as *Salmonella* and *Shigella*. Some *E. coli* strains, however, are human pathogens. These are

designated enterotoxigenic or enteropathogenic *E. coli*, respectively, based upon whether they produce toxins or whether they can invade the body through the gastrointestinal tract.

Enterotoxin-producing strains of *Escherichia coli* are also capable of causing both mild and severe forms of gastroenterocolitis. In most cases, enterotoxin-producing strains of *E. coli* do not invade the body through the gastrointestinal tract; rather, toxin released by cells growing on the surface lining of the gastrointestinal tract causes diarrhea. Aside from diarrhea, abdominal cramps are normally the only other clinical symptom of this disease. Travelers from the United States to Mexico often suffer severe diarrhea as a result of ingestion of strains of *E. coli* foreign to their own microbiota and therefore generally avoid drinking the water. Many cases of severe diarrhea in children are caused by noninvasive, enterotoxin-producing strains of *E. coli*. In some cases, enteropathogenic strains of *E. coli* invade the body through the mucosa of the large intestine to cause a serious form of dysentery. Invasive strains of *E. coli* are primarily associated with contaminated food and water in Southeast Asia and South America. The ability to invade the mucosa of the large intestine depends on the presence of a specific K antigen in enteropathogenic serotypes of *E. coli*. The enterotoxins produced by *E. coli* cause a loss of fluids from intestinal tissues. With proper replacement of body fluids and maintenance of the essential electrolyte balance, infections with enterotoxic *E. coli* normally are not fatal.

E. coli O157:H7 has emerged as an important foodborne and waterborne pathogen. This strain of *E. coli* causes hemorrhagic colitis and serious kidney disease. It frequently is fatal. Outbreaks of disease caused by infections with *E. coli* O157:H7 have most often been traced to contaminated undercooked hamburger meat. The U.S. Food and Drug Administration recommends thorough cooking of hamburger meat. Improved surveillance of meats for bacterial contamination is being implemented by the FDA.

Salmonella arizona, which formerly was known as *Arizona hinshawii* and which has been reclassified as a bioserotype of *S. enteritidis*, is associated with poultry, and products containing eggs such as custards. It causes gastroenteritis that generally lasts a few days. *Aeromonas hydrophila* has been the source of some foodborne disease outbreaks in the United States. It is ubiquitous in water and can infect various animals, including frogs. *Plesiomonas shigelloides* has been implicated in several foodborne disease outbreaks involving fish and shellfish. It can grow on minimal media containing ammonium

salts as the nitrogen source and glucose as the carbon source. Infections cause gastrointestinal upsets.

Tuberculosis, which is caused by *Mycobacterium tuberculosis* and related mycobacterial species, can be transmitted by the ingestion of contaminated food. Before the extensive use of pasteurization, milk contaminated with *M. tuberculosis* was associated with outbreaks of this disease. The dairy industry routinely screens milk for the presence of mycobacteria, such as *M. bovis* that commonly infects cattle and can also cause human disease.

Legionella species from potable waters can cause human infections when aerosols are inhaled. Most outbreaks of Legionnaire's disease have been traced to air-conditioning cooling systems. *Legionella* bacteria multiply in the cooling system waters, which are rapidly evaporated to provide cooling, and inadvertently become airborne and circulate through the air-conditioning system. In other cases potable waters, including aerosols from dental instruments and shower heads, are the source of *Legionella* infections. There have also been outbreaks of Legionnaire's disease due to contaminated aerosols used to moisten produce and keep fruits and vegetables fresh.

Mycotoxins produced by some fungi are responsible for serious cases of food poisoning. Many mycotoxins are potent neurotoxins. Various species of mushrooms contain toxins that can be absorbed through the gastrointestinal tract, and the ingestion of poisonous mushrooms, such as *Amanita phalloides*, is normally fatal. The amatoxins and phallotoxins produced by *A. phalloides* and other species of *Amanita* cause symptoms of food poisoning 8 to 24 hours after their ingestion. Initial symptoms include vomiting and diarrhea; later, degenerative changes occur in liver and kidney cells, and death may ensue within a few days of ingesting as little as 5 to 10 mg of toxin.

Some filamentous fungi, other than mushrooms, also produce toxins that can cause human disease. *Aspergillus* species growing on peanuts and grains produce aflatoxins, which are potent carcinogens, as well as toxic. They are known to cause death in sheep and cattle and may be involved in some human disease conditions. Aflatoxins are the only known carcinogens for which the United States government has set permissible levels; all other products with carcinogenic activity are banned outright.

Ergotism, another disease caused by fungi, results from ingesting grain containing ergot alkaloids produced by *Claviceps purpurea*. The toxins of *C. purpurea* cause degeneration of the capillary blood vessels, and this type of food poisoning has a relatively high mortality rate. Symptoms of ergotism may include vomiting, diarrhea, thirst, hallucinations,

convulsions, and lesions of the extremities. Various outbreaks of mass hallucinations have been traced to contamination of food with ergot alkaloids, and there are even theories that the Salem witch hunts in colonial Massachusetts were related to grain contamination and widespread ergotism.

Algae are rarely considered as the etiological agents of disease, but paralytic shellfish poisoning is caused by toxins produced by dinoflagellate *Gonyaulax*. Blooms of *Gonyaulax* cause red tides in coastal marine environments. During such algal blooms, the algae and the toxins they produce can be concentrated in bivalve shellfish, such as clams and oysters. The ingestion of shellfish containing algal toxins can lead to symptoms that resemble those of botulism. Shellfishing is banned in areas of *Gonyaulax* blooms to prevent this form of food poisoning.

Amebic dysentery or amebiasis is caused by the protozoan *Entamoeba histolytica*. Infections with *E. histolytica* may be asymptomatic or may involve mild or severe diarrhea and abdominal pain. Amebic dysentery occurs as a result of inadequate sewage treatment and contamination of water with *E. histolytica*, whose cysts are not killed by the chlorination methods normally used to treat municipal drinking water. Infection is acquired by ingesting contaminated food or water containing cysts of *E. histolytica*.

Giardia lamblia, a flagellated protozoan, is responsible for most cases of diarrhea infection caused by protozoa. The cysts of *G. lamblia* can enter the gastrointestinal tract through contaminated water. A high incidence of giardiasis occurred among groups touring Leningrad during the 1970s as a result of contaminated water supplies. In 1973 a major outbreak of giardiasis occurred in upstate New York, with an estimated 4,800 individuals developing symptoms of the disease. The following year, giardiasis was the most common waterborne disease in the United States. *G. lamblia* can live saprophytically within the small intestine without causing any symptoms of giardiasis, and in the United States almost 4% of the population appears to be infected by this organism. Excessive growth of the organism, however, can cause disease symptoms that include diarrhea, dehydration, mucus secretion, and flatulence.

Cryptosporidium is a newly emerging pathogen that has caused major outbreaks of waterborne disease in the United States. A 1993 outbreak of cryptosporidiosis in Milwaukee infected over 400,000 individuals. Excessive growth of this protozoan in municipal water supplies is the source of infection. New surveillance requirements are being implemented to prevent future major outbreaks of *Cryptosporidium* caused disease.

Food Spoilage

The growth of microorganisms in foods can alter the quality of the product, causing sizable economic losses to the food industry due to food spoilage. Food spoilage is a change in a food that renders it undesirable or unsafe for human consumption, and the growth of microorganisms in a food represents only one process that may cause food to spoil. The growth of pathogenic microorganisms in a food is certainly undesirable, as it can make that food unsafe to eat, but other microbially induced changes in food, such as decreased nutritional content and altered taste, odor, color, and texture, can also make a food undesirable for human consumption. Such food spoilage is difficult to define because it depends on the culture of the consumer; for example, Eskimos bury fish to produce a stinky mess and in Britain meat should have the strong aroma of aging, but these same foods would likely be rejected as spoiled in most parts of the United States.

Foods are classified according to their susceptibility to microbial spoilage as nonperishable, semiperishable, or perishable. Perishable foods, such as meats, fish, poultry, most fruits and vegetables, eggs, and milk, readily spoil because of microbial activities and generally have short shelf lives unless steps are taken to remove, kill, or prevent the growth of associated microorganisms. Semiperishable foods, such as potatoes and apples, generally remain unspoiled for prolonged periods of time unless improperly handled. Nonperishable foods, such as sugar, flour, and numerous dry products, normally do not spoil, but even these foods can spoil under appropriate conditions. For example, if cereals, grains, and flours are stored under conditions of high moisture, various fungal and bacterial populations are able to grow and spoil the products. We have all observed the growth of fungi on bread that has been stored too long.

The changes that occur in a food during microbial spoilage depend on the particular microbial populations involved, their enzymatic activities, environmental conditions, and the nature of the food. The microorganisms involved in the spoilage process generally do not originate within the food, the inner tissues of most plants and animals being sterile, but rather come from the surface tissues or are a result of contamination during processing. Several factors controlling food spoilage are intrinsic to the food, but others are extrinsic environmental parameters. The most important extrinsic factors influencing food spoilage are (1) temperature of storage; (2) relative humidity; and (3) oxygen concentration. By controlling these parameters, it is possible to alter the rate of food spoilage. It is somewhat more diffi-

cult to control the inherent properties of animal and plant tissues that influence microbial spoilage of foods. The intrinsic moisture content, pH, and physical and chemical nature of the food, in part, control the numbers and types of microorganisms involved in the spoilage process. Fruits, vegetables, meats, poultry, seafoods, milk and dairy products, and various other food products differ in their biochemical composition and therefore are subject to spoilage by differing microbial populations.

Fruits and vegetables are subject to rot because of the microbial degradation of pectin, the biochemical responsible for maintaining the firmness and texture of fruits and vegetables. Microbially produced pectinesterases and polygalacturonases hydrolyze pectins, resulting in the formation of soft spots in fruits and vegetables. About 20% of the harvested crops of fruits and vegetables are lost to spoilage primarily because of the activities of bacteria and fungi. Carbohydrates, which are present in high concentrations in fruits, vegetables, and other foods, are readily degraded by numerous microorganisms, resulting in the production of various degradation products, such as low molecular weight acids and alcohols. The accumulation of such products of microbial metabolism can alter the taste of a food, generally causing it to sour. The taste of sour milk, for example, is associated with the accumulation of lactic acid from the microbial transformation of carbohydrates. The accumulation of low molecular weight acids may also alter the smell of the product; sour milk often can be recognized just by smelling it. In canned foods, production of acid and no gas is referred to as flat-sour spoilage because the food becomes sour, but the can shows no evidence of food spoilage because no gas is produced; that is, the can remains flat. When gas is produced during spoilage of canned foods, the can swells, as can be seen by examining the lids, which are designed to push outward if pressure builds within the can because of microbial action. Thermophilic microorganisms, that is, those that can withstand exposure to elevated temperatures, and especially endospore formers, are important in the spoilage of canned foods.

Meats and other proteinaceous products can be decomposed by anaerobic bacteria, resulting in putrefaction. Putrefaction of meat is the result of the breakdown of proteins by proteinases. The subsequent degradation of amino acids produces foul-smelling, low molecular weight sulfur- and nitrogen-containing compounds, such as mercaptans, hydrogen sulfide, ammonia, and amines. The evolution of noxious, odoriferous compounds from putrefying proteins renders food unacceptable for human consumption. The characteristic odor of hydrogen sul-

fide, for example, renders rotting eggs inedible, and the development of off odors and slime on poultry and beef reflects the presence of increased microbial populations and spoilage of the meat. In canned foods such spoilage is referred to as sulfide stinkers. Under aerobic conditions, the decomposition of proteins generally does not result in the production of compounds with obnoxious odors. The spoilage of meat under aerobic conditions, though, can result in the accumulation of surface slime, generally because of the growth of *Pseudomonas*, *Achromobacter*, *Streptococcus*, *Leuconostoc*, *Bacillus*, *Micrococcus*, and *Lactobacillus* species. The physical state of the meat influences which microbial species may be involved in spoilage. The spoilage of fresh whole meats is normally associated with lactic acid bacteria, particularly *Lactobacillus*, *Leuconostoc*, and *Streptococcus* species, whereas the spoilage of ground beef is primarily due to the growth of *Pseudomonas*, *Achromobacter*, and *Micrococcus*.

Spoilage by microbial degradation of fats and oils produces rancidity, caused by oxidation or hydrolysis of lipids. Spoiled butter, for example, becomes rancid because of the hydrolysis of butterfat, with the production of free fatty acids and glycerol and the accompanying development of undesirable flavors. Fish with high lipid levels, such as salmon and mackerel, may also become rancid because of the hydrolysis of fats and oils.

The growth of some microorganisms can alter the color of the food. For example, the growth of *Serratia marcescens* can produce bright red bread; the combined growth of *Pseudomonas* species and *Streptococcus lactis* produces blue milk; the growth of fungi on meat surfaces can produce off colors, such as black spot caused by *Cladosporium herbarum*, white spot caused by *Sporotrichium carnis*, and green patches produced by *Penicillium* species; and the growth of heterolactic fermenters on cured meats, such as frankfurters, causes greening because of the action of peroxidases on the meat pigments. The development of unnatural colors in a food reduces its acceptability, regardless of whether there is any associated development of an off taste or smell. Microbial growth can also alter the texture of a food. For example, the growth of *Bacillus* species in products such as milk and dough can produce ropiness, a textural change resulting from the growth of encapsulated *Bacillus* species together with the hydrolysis of starch and proteins.

Microbiological Production of Food

Although microbial growth is a problem when it results in food spoilage, microorganisms are used beneficially in the food industry for food production.

Many of the foods and beverages we commonly enjoy, such as wine and cheese, are the products of microbial enzymatic activity. For the most part, it is the fermentative metabolism of microorganisms that is exploited in the production of food products. The accumulation of fermentation products, such as ethanol and lactic acid, is desirable because of their characteristic flavors and other properties. Only a few processes, such as the production of vinegar, make use of microbial oxidative metabolism. The microbial production of foods can be viewed as an exercise in harnessing microbial biochemistry to produce desired, rather than adverse, changes in food products.

The production of fermented foods requires the proper substrates, microbial populations, and environmental conditions to obtain the desired end product. Quality control is essential in food fermentation to ensure that the product is of high quality. A fermented food may require additional preservation to prevent spoilage because further uncontrolled microbial growth could render it inedible. For example, once wine is produced, it must be maintained under anaerobic conditions in order to prevent its oxidation to vinegar.

The microbial processes used in food production traditionally employ microbial enzymatic activities to transform one food into another, with the microbially produced food product having properties vastly different from those of the starting material. In addition to the use of microorganisms to produce fermented food products, microbial biomass is now considered a potential source of protein for meeting the food needs of an expanding world population. Some microorganisms, such as mushrooms, have been used as food products for centuries. The growth of bacteria, algae, and fungi as proteinaceous food, however, is a relatively new concept. Microbial biomass can be used as an animal feed supplement or may be developed as a direct source of protein for human consumption.

Numerous products are made by the microbial fermentation of milk, including buttermilk, yogurt, and many cheeses. The fermentation of milk is primarily carried out by lactic acid bacteria. The lactic acid fermentation pathway and the accumulation of lactic acid from the metabolism of the milk sugar lactose are common to the production of fermented dairy products. The accumulated lactic acid in these products acts as a natural preservative. The differences in the flavor and aroma of the various fermented dairy products are due to additional fermentation products that may be present in only relatively low concentrations.

Different fermented dairy products are produced by using different strains of lactic acid bacteria as

starter cultures and different fractions of whole milk as the starting substrate. Sour cream, for example, uses *Streptococcus cremoris* or *S. lactis* for the production of lactic acid, and *Leuconostoc cremoris* or *S. lactis diacetylactis* for the production of the characteristic flavor compounds. Cream is the starting substrate for this product. If skim milk is used as the starting material, cultured buttermilk is produced. Bulgarian buttermilk is made by using *Lactobacillus bulgaricus* for the production of both lactic acid and flavor compounds. Butter is normally made by churning cream that has been soured by lactic acid bacteria. *S. cremoris* or *S. lactis* is used to produce lactic acid rapidly, and *Leuconostoc citrovorum* produces the necessary flavor compounds. The *Leuconostoc* enzymes attack citrate in milk, producing diacetyl, which gives butter its characteristic flavor and aroma. Kefir and koumis, which are popular in some European countries, are fermentation products of *S. lactis*, *S. cremoris*, other *Lactobacillus* species, and yeasts. Lactic acid, ethanol, and carbon dioxide are formed during the fermentation and gives these products their characteristic flavors.

Yogurt is made by fermenting milk with a mixture of *L. bulgaricus* and *S. thermophilus*. The characteristic flavor of yogurt is due to the accumulation of lactic acid and acetaldehyde produced by *L. bulgaricus*. Because of the tart taste of acetaldehyde, most yogurt produced in the United States is flavored by adding fruit.

A wide variety of cheeses is produced by microbial fermentation. Cheeses consist of milk curds that have been separated from the liquid portion of the milk (whey). The curdling of milk is accomplished by using the enzyme rennin (casein coagulase or chymosin) and lactic acid bacterial starter cultures. Rennin is obtained from calf stomachs or by microbial production. The natural production of cheeses involves lactic acid fermentation, with various mixtures of *Streptococcus* and *Lactobacillus* species used as starter cultures to initiate the fermentation. The flavors of different cheeses result from the use of different microbial starter cultures, varying incubation times and conditions, and the inclusion or omission of secondary microbial species late in the fermentation process.

Ripening of cheeses involves additional enzymatic transformations after the formation of the cheese curd, using enzymes produced by lactic acid bacteria or enzymes from other sources. Unripened cheeses do not require the additional enzymatic transformations. Cottage cheese and cream cheese are produced by using a starter culture similar to the one used for the production of cultured buttermilk and are soft

cheeses that do not require ripening. Sometimes a cheese is soaked in brine to encourage the development of selected bacterial and fungal populations during ripening. Limburger is a soft cheese produced in this manner. During ripening the curds are softened by proteolytic and lipolytic enzymes, and the cheese acquires its characteristic aroma. The production of Parmesan cheese also involves brine curing.

Swiss cheese formation involves a late propionic acid fermentation, with ripening accomplished by *Propionibacterium shermanii* and *P. freudenreichii*. The propionic acid yields the characteristic aroma and flavor, and the carbon dioxide produced during this late fermentation forms the holes in Swiss cheese. Various fungi are also used in the ripening of different cheeses. The unripened cheese is normally inoculated with fungal spores and incubated in a warm, moist room to promote the growth of filamentous fungi. For example, blue cheeses are produced by using *Penicillium* species. Roquefort cheese is produced by using *P. roqueforti*, and Camembert and Brie by using *P. camemberti* and *P. candidum*.

Several types of sausage, such as Lebanon bologna, the salamis, and the dry and semidry summer sausages, are produced by allowing the meat to undergo heterolactic acid fermentation during curing. The fermentation has a preservative effect and also adds a tangy flavor to the meat. Various lactic acid bacteria are normally involved in the fermentation, but *Pediococcus cerevisiae* can be used for controlled production of these types of meats.

Yeasts are added to bread dough to ferment the sugar, producing the carbon dioxide that leavens the dough and causes it to rise. The principal yeast used in bread baking is *Saccharomyces cerevisiae*, known as bakers' yeast. Bakers' yeast is produced in large quantities for the baking industry. The yeast is normally grown in a molasses-mineral salts medium at a pH of 4.3 to 4.5 and temperature of 30°C, with the molasses substrate added gradually to maintain a sugar concentration of 0.5 to 1.5%. The concentration of sugar in the fermentor is critical, as too high a concentration represses respiratory enzymes and alcohol production even under highly aerobic conditions. The yeasts are generally collected by centrifugation and pressed through a filter to remove excess liquid. For the baking industry, the yeasts are normally formed into cakes or are dried further to form active dry yeast containing less than 8% water and are frequently used for home baking purposes.

In the baking process, the yeast is used strictly as a source of enzymes to carry out alcoholic fermentation. The yeast does not grow during the first 2 hours after addition to the dough, by which time the leav-

ening process is normally completed. Amylases in the dough convert starch to sugars, and the yeasts metabolize the sugars that are formed, producing carbon dioxide and ethanol. Besides *S. cerevisiae*, various other microorganisms, including coliform bacteria and *Clostridium* species, can be employed for leavening bread. The microorganisms used for bread leavening must produce carbon dioxide from the fermentation of sugars to be useful.

In addition to leavening bread, microbes produce the characteristic flavors of some breads. For example, the production of San Francisco sourdough bread utilizes the yeast *Torulopsis holmii* and a heterofermentative *Lactobacillus* species to sour the dough and give this bread its characteristic sour flavor. Rye bread is also produced by initially souring the dough; cultures of *Lactobacillus plantarum*, *L. brevis*, *L. bulgaricus*, *Leuconostoc mesenteroides*, and *Streptococcus thermophilus* are employed as starter cultures in making different rye breads. The action of heterofermentative lactic acid bacteria produces the bread's characteristic flavor.

Microorganisms, principally yeasts in the genus *Saccharomyces*, are used to produce various types of alcoholic beverages. The production of alcoholic beverages relies on the conversion of sugar to alcohol by microbial enzymes. The flavor and other characteristic differences between various types of alcoholic beverages reflect differences in the starting substrates and the production process, rather than differences in the microbial culture or the primary fermentation pathways employed in the production of alcoholic beverages.

The production of vinegar involves an initial anaerobic fermentation to convert carbohydrates by *S. cerevisiae* to alcohol, followed by a secondary oxidative transformation of the alcohol to form acetic acid by *Acetobacter* and *Gluconobacter*. The starting materials for the production of vinegar may be fruits, such as grapes, oranges, apples, pears; vegetables, such as potatoes; malted cereals, such as barley, rye, wheat, and corn; and sugary syrups such as molasses, honey, and maple syrup. The type of vinegar is determined by the starting material. For example, wine vinegar comes from grapes and cider vinegar from other fruits. The history of the commercial production of vinegar shows an interesting progression in fermentor design to accomplish the necessary transfer of oxygen to the bacteria, and we will examine some of the types of vinegar generators that have been developed.

Vegetables, such as cabbages, carrots, cucumbers, green tomatoes, leafy vegetables, greens, and olives, are fermented by using lactic acid bacteria as a means

of creating new food products that are not readily subject to spoilage. Other fermentations, particularly of soybeans, are carried out to produce specially desired flavors, aromas, and textures in food products.

The production of sauerkraut involves a succession of bacterial populations. Coliform bacteria, such as *Enterobacter cloacae*, are prominent in the initial mixed community, and produce gas and volatile acids as well as some lactic acid. The accumulating lactic acid exerts a selective pressure on the microbial community, causing population shifts and continued succession. As a result, after the initial fermentation there is a shift in the composition of the microbial community, and *Leuconostoc mesenteroides*, which grows well at 21°C and is not inhibited by 2.5% salt, becomes the dominant microbial population. Up to 1% lactic acid may accumulate—and yeasts and various bacteria may grow as a surface film—during this phase of the fermentation. The continuing succession of bacterial populations next favors the development of *Lactobacillus plantarum*, which produces acid but no gas. During this phase of the fermentation, the concentration of lactic acid reaches 1.5 to 2%. Growth of *L. plantarum* also removes mannitol, which is produced by *Leuconostoc* and has an undesirable bitter flavor. The fermentation can be stopped at this stage by canning or refrigerating the sauerkraut.

The traditional method for producing pickles by fermenting cucumbers uses the natural microbiota associated with the cucumber, and controlled temperature and salt concentrations, to regulate the fermentation process. Controlled fermentation of pickles can be achieved by inoculation with *Lactobacillus plantarum* and *Pediococcus cerevisiae*. At the beginning of the fermentation, when the salt concentration is low, many bacterial genera are able to grow, including *Pseudomonas*, *Flavobacterium*, and *Bacillus*. As the salt concentration is increased, the populations that become favored include the lactic acid bacteria *Leuconostoc mesenteroides*, *Streptococcus faecalis*, and *P. cerevisiae*. As the lactic acid and salt concentrations increase, *L. plantarum* becomes the dominant bacterium, beginning several days after the fermentation and continuing until the salt concentration surpasses 10%. The completion of the fermentation process involves yeasts that grow at high salt concentrations. During the final yeast fermentation stage, some carbohydrates are converted to alcohol. The growth of film-forming yeasts, such as *Debaryomyces*, *Pichia*, *Endomycopsis*, and *Candida*, lowers the lactic acid concentration.

Because of the complexity of changes in the microbial community during this natural fermentation, the

process often goes awry and yields unmarketable pickles, such as (1) floaters and bloaters that float because of excessive gas accumulation within the cucumber; (2) hollow pickles, in which the cucumber contents have shriveled because of excessive salt or the formation of high concentrations of acetic acid; (3) stinkers, due to the accumulation of H_2S ; (4) black pickles, due to bacterial pigment production; (5) soft pickles, due to fungal proteases; and (6) slippery pickles, due to the surface growth of encapsulated bacteria. Controlled fermentation conditions—and a pure inoculum of *P. cerevisiae* and *L. plantarum* after the removal of the natural microbiota by fumigation or chlorination—can be used to increase the likelihood of producing a quality pickle.

The production of green olives involves lactic acid fermentation. The harvested olives are washed with a solution of sodium hydroxide that removes most of the oleuropein, a very bitter phenolic glucoside that gives unfermented olives a very undesirable flavor. The olives are then placed in a brine solution, and a lactic acid fermentation lasting for 6 to 10 months is permitted to occur. During the first 2 weeks of the fermentation, the brine becomes stabilized as compounds are leached from the olives and microbial populations begin to multiply. At the intermediate stage, which occurs during the following 2 to 3 weeks, *Leuconostoc* is the dominant bacterial species and lactic acid accumulates. The final stage of fermentation is dominated by *L. plantarum* and *L. brevis*; yeasts and various bacteria also occur during this stage.

Several oriental foods are prepared by fermenting soybeans or rice. Soy sauce, a brown, salty, tangy sauce, which in Japanese is called *shoyu*, is produced from a mash consisting of soybeans, wheat, and wheat bran. Soy sauce is used as a condiment or as an ingredient in other sauces. The starter culture for the production of soy sauce is produced by koji fermentation, a dry fermentation in which a mixture of soybeans and wheat is inoculated with spores of *Aspergillus oryzae*. The mixture is moistened but is not submerged in liquid. The fungi grow on the surface of the soybeans and wheat, accumulating various enzymes including proteinases and amylases. Various bacterial populations, normally dominated by lactic acid bacteria, also develop during this koji fermentation. After the starter culture develops, it is dried and extracted. The maturation of soy sauce begins with lactic acid bacteria, including lactic acid production by *Pediococcus soyae*, and later involves

alcoholic fermentations by yeasts, such as *Saccharomyces rouxii*, *Zygosaccharomyces soyae*, and *Torulopsis* species. The most important organisms during the fermentation process are *A. oryzae*, which produces proteinases and amylases; *Lactobacillus* species, which produce sufficient amounts of lactic acid to prevent spoilage by other microorganisms; and yeasts, which produce sufficient alcohol to increase the flavor.

Miso is also produced by using a koji fermentation with *A. oryzae*. Steamed polished rice, placed in shallow trays, is used in the production of the starter culture. The koji is mixed with a mash of steamed soybeans and after the addition of salt, the fermentation is allowed to proceed at 28°C for 1 week, at 35°C for 2 months, and at room temperature for several additional weeks. The miso is normally ground into a paste, to be combined with other food before eating.

Tempeh is an Indonesian food produced from soybeans. The soybeans are soaked at 25°C, dried, and inoculated with spores of various species of *Rhizopus*. The mash is incubated at 32°C for 20 hours, during which mycelial growth occurs. The product is then salted and fried before eating.

Tofu (Japanese) or sofu (Chinese) is a cheese-like product produced by fermenting soybeans with *Mucor* species. The soybeans are soaked, ground to a paste, and curdled by adding calcium or magnesium salts. The pressed curd blocks are placed in trays at 14°C and incubated for 1 month, during which time the fungal populations develop.

Natto also is produced from boiled soybeans and involves the incubation of *Bacillus subtilis* with soybeans for 1 to 2 days, during which time proteinase enzymes soften and add flavor compounds to the soybeans. Various other oriental foods are also produced by similar fermentations.

Poi is a fermented food product from the Hawaiian Islands. In the production of poi, the stems of the taro plant are steamed, ground, and subjected to fermentation for 1 to 6 days. During the first few hours, coliforms, *Pseudomonas*, and various other microorganisms predominate. Then a successional process occurs, with *Lactobacillus*, *Streptococcus*, and *Leuconostoc* becoming the dominant populations. Finally, yeasts and the fungus *Geotrichum candidum* flourish. The fermentation products, principally lactic acid, acetic acid, formic acid, ethanol, and carbon dioxide, contribute to the characteristic texture, flavor, and aroma of poi.

A-1 Medium (BAM M1)

Composition per liter:

Pancreatic digest of casein	20.0g
Lactose	5.0g
NaCl	5.0g
Salicin	0.5g
Triton™ X-100	1.0mL
pH 6.9 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.9. Distribute into test tubes containing an inverted Durham tube. Medium may be cloudy prior to autoclaving. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the detection of fecal coliforms in foods and waters by a most-probable-number (MPN) method. Multiple dilutions of samples (3, 5, or 10 replicates per dilution) are added to tubes containing A-1 medium. After incubation, test tubes with gas accumulation in the Durham tubes are scored positive and those with no gas as negative. A MPN table is consulted to determine the most probable number of fecal coliforms.

AAM Medium (DSMZ Medium 57)

Composition per liter:

Casamino acids	7.0g
Yeast extract	7.0g
Tryptone	5.0g
Meat extract	5.0g
Na-acetate	2.5g
MgSO ₄ ·7H ₂ O	200.0mg
MnSO ₄ ·2H ₂ O	50.0mg
Tween™ 80	1.0mL
pH 5.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.4. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus* spp.

Abeyta-Hunt Bark Agar (BAM M29a)

Composition per 1016mL:

Beef heart, infusion from	500.0g
Agar	15.0g

Tryptose	10.0g
NaCl	5.0g
Yeast extract	2.0g
Horse blood, lysed	50.0mL
Cefoperazone solution	4.0mL
Rifampicin solution	4.0mL
Amphotericin B solution	4.0mL
Ferrous sulfate pyruvate metabisulfite solution	4.0mL
pH 7.4 ± 0.2 at 25°C	

Amphotericin B Solution:

Composition per 100mL:

Amphotericin B	0.05g

Preparation of Amphotericin B Solution:

Add Amphotericin B to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cefoperazone Solution:

Composition per 100mL:

Sodium cefoperazone	0.08g

Preparation of Cefoperazone Solution: Add sodium cefoperazone to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Rifampicin Solution:

Composition per 100mL:

Rifampicin	0.25g

Preparation of Rifampicin Solution: Add rifampicin to 70.0mL ethanol. Mix thoroughly. Add distilled/deionized water to bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Ferrous Sulfate, Pyruvate, Metabisulfite Solution:

Composition per 100mL:

FeSO ₄	6.25g
Na-pyruvate	6.25g
Na-metabisulfite	6.25g

Preparation of Ferrous Sulfate, Pyruvate, Metabisulfite Solution: Add Na-pyruvate to 20mL distilled/deionized water. Mix thoroughly. Add Na-metabisulfite and FeSO₄. Bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cefoperazone solution, amphotericin B solution, rifampicin solution, ferrous sulfate pyruvate metabisulfite solution, and horse blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 4.0mL cefoperazone solution, 4.0mL amphotericin B solution, 4.0mL rifampicin solution, 4.0mL ferrous sulfate pyruvate metabisulfite

fide solution, and 50.0mL lysed horse blood. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Campylobacter* spp.

ABY Agar (Acid Bismuth Yeast Agar)

Composition per liter:

Agar	20.0g
Glucose	20.0g
Bi ₂ (SO ₃) ₂	8.0g
(NH ₄) ₂ SO ₄	3.0g
KH ₂ PO ₄	3.0g
MgSO ₄ ·7H ₂ O	0.25g
CaCl ₂ ·2H ₂ O	0.25g
Biotin	10.0μg

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes in a slanted position.

Use: For the selective isolation and differentiation of *Candida albicans*, which can cause food allergies, from other *Candida* species. *Candida albicans* and *Candida tropicalis* colonies appear as smooth, brownish-black round colonies. Other *Candida* species are differentially pigmented or produce diffusible pigments. Usually used in conjunction with BiGGY agar to differentiate further *Candida*; on BiGGY agar, *Candida albicans* appears as brown to black colonies with no pigment diffusion and no sheen, whereas *Candida tropicalis* appears as dark brown colonies with black centers, black pigment diffusion, and a sheen.

AC Agar (AC Medium)

Composition per liter:

Proteose peptone No. 3	20.0g
Glucose	5.0g
Beef extract	3.0g
Yeast extract	3.0g
Malt extract	3.0g
Ascorbic acid	0.2g
Agar	1.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and isolation of anaerobes, microaerophiles, and aerobes. Recommended for the sterility testing of solutions and other materials not containing mercurial preservatives.

AC Broth

Composition per liter:

Proteose peptone No. 3	20.0g
Glucose	5.0g
Beef extract	3.0g
Yeast extract	3.0g
Malt extract	3.0g
Ascorbic acid	0.2g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and isolation of a wide variety of microorganisms, including anaerobes, microaerophiles, and aerobes. Recommended for the sterility testing of solutions and other materials not containing mercurial preservatives.

Acetamide Agar

Composition per liter:

Agar	15.0g
Acetamide	10.0g
NaCl	5.0g
K ₂ HPO ₄	1.0g
NH ₄ H ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.2g
Bromthymol Blue	0.08g

pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes in a slanted position to produce a long slant.

Use: For the differentiation of nonfermentative Gram-negative bacteria, especially *Pseudomonas aeruginosa*. Bacteria that deaminate acetamide turn the medium blue.

Acetamide Agar

Composition per liter:

Agar	15.0g
Acetamide	10.0g
NaCl	5.0g
K ₂ HPO ₄	1.39g
KH ₂ PO ₄	0.73g
MgSO ₄ ·7H ₂ O	0.5g
Phenol Red	0.012g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes in a slanted position to produce a long slant.

Use: For the differentiation of nonfermentative Gram-negative bacteria, especially *Pseudomonas aeruginosa*. Bacteria that deamidate acetamide turn the medium blue.

Acetamide Broth

Composition per liter:

Acetamide	10.0g
NaCl	5.0g
K ₂ HPO ₄	1.39g
KH ₂ PO ₄	0.73g
MgSO ₄ ·7H ₂ O	0.5g
Phenol Red	0.012g

pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of nonfermentative Gram-negative bacteria, especially *Pseudomonas aeruginosa*. Bacteria that deamidate acetamide turn the broth purplish red.

Acetamide Medium (BAM M2)

Composition per liter:

Stock basal solution	400.0mL
Stock acetamide solution	100.0mL

pH 6.9 ± 0.2 at 25°C

Stock Basal Solution:

Composition per 400.0mL:

Agar	0.5g
KH ₂ PO ₄ solution, 0.5M	14.0mL
K ₂ HPO ₄ solution, 0.5M	6.0mL

Preparation of Stock Basal Solution: Add components, except PR-CV solution, to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly. Gently heat and bring to boiling with agitation to dissolve agar. Add 1.0mL PR-CV solution.

PR-CV Solution:

Composition per 200.0mL:

Phenol Red	2.0g
Crystal Violet	0.2g

Preparation of PR-CV Solution: Add components to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Add 5N NaOH while stirring until components are dissolved.

Stock Acetamide Solution:

Composition per 100.0mL:

Acetamide	1.0g

Preparation of Stock Acetamide Solution:

Add acetamide to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Store over methylene chloride in a screw capped container. Can be stored indefinitely at room temperature.

Preparation of Medium: Combine 400.0mL stock basal solution and 100.0mL stock acetamide solution. Mix thoroughly. Distribute into tubes or flasks. Steam for 10 min at 100°C. Cool.

Use: For the differentiation of nonfermentative Gram-negative bacteria, especially *Pseudomonas aeruginosa*, e.g., in milk.

Acetate Agar (BAM M3)

Composition per liter:

Agar	20.0g
NaCl	5.0g
Sodium acetate	2.0g
KH ₂ PO ₄	1.0g
(NH ₄) ₂ PO ₄	1.0g
MgSO ₄	0.2g
Bromthymol blue	0.08g

pH 6.7 ± 0.2 at 25°C

Preparation of Medium: Add components, except MgSO₄, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.7. Add 0.2g MgSO₄. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically distribute into tubes or flasks. For slants distribute aliquots into tubes prior to autoclaving. After autoclaving allow tubes to cool in inclined position to obtain a 5cm slant.

Use: For the cultivation of *Leuconostoc* species.

Acetate Broth**Composition per liter:**

Meat extract	50.0g
Glucose	10.0g
Peptone.....	5.0g
Yeast extract.....	5.0g
Sodium acetate buffer	100.0mL
Tween 80.....	0.5mL

pH 5.4 ± 0.2 at 25°C

Sodium Acetate Buffer:**Composition per liter:**

Sodium acetate·3H ₂ O.....	272.2g

Preparation of Sodium Acetate Buffer: Add sodium acetate to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.4 with glacial acetic acid. Filter sterilize.

Preparation of Medium: Add components, except sodium acetate buffer, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 5.4. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile sodium acetate buffer. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and cultivation of *Leuconostoc* species and *Pediococcus* species.

Acetate Differential Agar**(Sodium Acetate Agar)****(Simmons' Citrate Agar, Modified)****Composition per liter:**

Agar	20.0g
NaCl.....	5.0g
Sodium acetate.....	2.0g
(NH ₄)H ₂ PO ₄	1.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
Bromthymol Blue	0.08g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to cold distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes to produce a 1 cm butt and 30 cm slant. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes in a slanted position.

Use: For the differentiation of *Shigella* species from *Escherichia coli* and also for the differentiation of nonfermenting Gram-negative bacteria. Bacteria that can utilize acetate as the sole carbon source turn the medium blue.

Acetobacter Agar**Composition per liter:**

Agar	15.0g
Yeast extract.....	5.0g
(NH ₄) ₂ SO ₄	3.3g
KH ₂ PO ₄	1.0g
MgSO ₄	0.25g
Vitamin solution.....	200.0mL
Glucose solution	15.0mL
Trace elements solution	1.0mL

Trace Elements Solution:**Composition per 100.0mL:**

CaCl ₂ ·2H ₂ O	1.457g
FeSO ₄ ·7H ₂ O.....	0.366g
ZnSO ₄ ·7H ₂ O.....	0.178g
MnSO ₄ ·H ₂ O.....	0.101g
Na ₂ MoO ₄ ·2H ₂ O.....	23.4mg
CuSO ₄ ·5H ₂ O.....	7.8mg

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Vitamin Solution:**Composition per 100.0mL:**

<i>m</i> -Inositol.....	200.0mg
Calcium DL-pantothenate.....	40.0mg
Nicotinic acid.....	40.0mg
Pyridoxine·HCl.....	40.0mg
Thiamine·HCl	40.0mg
<i>p</i> -Aminobenzoic acid.....	20.0mg
Riboflavin	20.0mg
Biotin	0.2mg
Folic acid	0.2mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Glucose Solution:**Composition per 100.0mL:**

Glucose	40.0g

Preparation of Glucose Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except vitamin solution, glucose solution, and trace element solution, to distilled/deionized water and bring volume to 784.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 200.0mL of sterile vitamin solution, 15.0mL of sterile glucose solution, and 1.0mL of sterile trace elements solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Acetobacter xylinum*.

***Acetobacter/Gluconobacter* Agar**

Composition per liter:

Glucose	100.0g
Agar	25.0g
CaCO ₃	20.0g
Yeast extract	10.0g

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Acetobacter aceti*, *Acetobacter liquefaciens*, *Acetobacter pasteurianus*, *Acetobacter xylinum*, *Frateuria aurantia*, and *Gluconobacter oxydans*.

***Acetobacter* Medium**

Composition per liter:

Agar	15.0g
Autolyzed yeast	10.0g
CaCO ₃	10.0g
Glucose	3.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes to produce a 1cm butt and 30cm slant. Autoclave for 15 min at 15 psi pressure–121°C. Agitate tubes to mix CaCO₃. Cool tubes rapidly in a slanted position to keep the CaCO₃ in suspension.

Use: For the cultivation and maintenance of *Acetobacter* species and *Gluconobacter* species.

***Acetobacter xylinum* Medium**

Composition per liter:

Glucose	50.0g
Yeast extract	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Acetobacter xylinum*.

Acetogen Medium

Composition per 421.8mL:

NaHCO ₃	2.4g

NH ₄ Cl	0.2g
Yeast extract	0.2g
Stock salts solution #1	40.0mL
Potassium phosphate buffer	20.0mL
Clarified rumen fluid	20.0mL
Stock salts solution #2	4.0mL
Trace minerals	4.0mL
Vitamin solution	4.0mL
Reducing agent	4.0mL
Tungstate solution	0.4mL
Resazurin (0.1% solution)	0.4mL

Potassium Phosphate Buffer:

Composition per 830.0mL:

K ₂ HPO ₄	15.68g
KH ₂ PO ₄	4.72g

Preparation of Potassium Phosphate Buffer:

Dissolve K₂HPO₄ in 600.0mL of distilled/deionized water and KH₂PO₄ in 230.0mL of distilled/deionized water. Mix the two solutions together and use.

Stock Salts Solution #1:

Composition per liter:

KCl	1.6g
NaCl	1.4g
MgSO ₄ ·7H ₂ O	0.2g

Preparation of Stock Salts Solution #1: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Stock Salts Solution #2:

Composition per liter:

CaCl ₂ ·2H ₂ O	0.1g

Preparation of Stock Salts Solution #2: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Trace Minerals:

Composition per liter:

Nitrilotriacetic acid	1.5g
MgSO ₄ ·7H ₂ O	3.0g
MnSO ₄ ·H ₂ O	0.5g
NaCl	1.0g
NiCl ₂ ·6H ₂ O	0.1g
FeSO ₄ ·7H ₂ O	0.1g
CoCl ₂ ·6H ₂ O	0.1g
CaCl ₂	0.1g
ZnSO ₄ ·7H ₂ O	0.1g
Na ₂ SeO ₃ ·5H ₂ O	0.01g
CuSO ₄ ·5H ₂ O	0.01g
AlK(SO ₄) ₂ ·12H ₂ O	0.01g
H ₃ BO ₃	0.01g
Na ₂ MoO ₄ ·2H ₂ O	0.01g

Preparation of Trace Minerals: Add nitrilotriacetic acid to 500.0mL of distilled/deionized water. Dissolve by adjusting pH to 6.5 with KOH. Bring

volume to 1.0L with distilled/deionized water. Add remaining components. Mix thoroughly.

Vitamin Solution:

Composition per liter:

Pyridoxine-HCl	10.0mg
Ascorbic acid	5.0mg
Calcium pantothenate	5.0mg
Choline chloride	5.0mg
Lipoic acid	5.0mg
<i>i</i> -Inositol	5.0mg
Niacinamide	5.0mg
Nicotinic acid	5.0mg
<i>p</i> -Aminobenzoic acid	5.0mg
Pyridoxal-HCl	5.0mg
Riboflavin	5.0mg
Thiamine-HCl	5.0mg
Biotin	2.0mg
Folic acid	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Store frozen.

Tungstate Solution:

Composition per liter:

Na ₂ WO ₄ ·2H ₂ O	99.0mg

Preparation of Tungstate Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Reducing Agent:

Composition per 110.0mL:

L-Cysteine·HCl·H ₂ O	2.5g
Na ₂ S·9H ₂ O	2.5g

Preparation of Reducing Agent: Add 110.0mL of distilled/deionized water to a 250.0mL round-bottomed flask. Boil under N₂ gas for 1 min. Cool to room temperature. Add L-cysteine·HCl and dissolve. Adjust to pH 9 with 5*N* NaOH. Add washed Na₂S·9H₂O and dissolve. Distribute in amounts needed into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except NaHCO₃ and reducing agent, to distilled/deionized water and bring volume to 417.8mL. Mix thoroughly. Gently heat and bring to boiling under 80% N₂ + 20% CO₂. Cool to 45°–50°C. Add NaHCO₃ and reducing agent. Distribute into tubes or flasks under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C. After inoculation, exchange headspace with 80% H₂ + 20% CO₂.

Use: For the cultivation and maintenance of acetogenic anaerobes such as some *Clostridium* species.

***Achromobacter* Choline Medium**

Composition per liter:

NaCl	30.0g
Agar	18.0g
Choline chloride	5.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
FeSO ₄ ·7H ₂ O	0.01g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix well and warm gently until dissolved. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Achromobacter* spp. and other bacteria that can utilize choline as a carbon source.

***Achromobacter* Choline Medium, Modified**

Composition per liter:

NaCl	30.0g
Agar	15.0g
Choline chloride	5.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	1.0g
FeSO ₄ ·7H ₂ O	0.018g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Peel and finely dice potatoes and carrots. Add to 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter solids through cheesecloth. Add MgSO₄·7H₂O, FeSO₄·7H₂O, and agar to filtrate. Mix thoroughly. Bring volume to 1.0L with distilled/deionized water. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Achromobacter cholinophagum*.

***Achromobacter* Medium (ATCC Medium 457)**

Composition per liter:

K ₂ HPO ₄	7.32g
Ammonium tartrate	4.6g
KH ₂ PO ₄	1.09g
MgSO ₄ ·7H ₂ O	0.04g
FeSO ₄ ·7H ₂ O	0.04g
CaCl ₂ ·2H ₂ O	0.014g
MgSO ₄ ·7H ₂ O	0.002g

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix well and warm gently until dissolved. Distribute into test tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Achromobacter* species and *Alcaligenes* species.

***Achromobacter* Medium (ATCC Medium 589)**

Composition per liter:

Agar	20.0g
K ₂ HPO ₄	7.0g
Methionine	5.0g
KH ₂ PO ₄	2.0g
(NH ₄) ₂ SO ₄	1.0g
Sodium citrate	0.4g
MgSO ₄ ·7H ₂ O	0.1g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Achromobacter* species.

Acid Broth (BAM M4)

Composition per liter:

Glucose	5.0g
Proteose peptone	5.0g
Yeast extract	5.0g
K ₂ HPO ₄	4.0g

pH 5.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation of bacteria from canned foods.

Acid Egg Medium

Composition per 1640.0mL:

Potato starch	30.0g
KH ₂ PO ₄	12.3g
Malachite Green	0.4g
MgSO ₄ ·7H ₂ O	0.3g
Penicillin G	100,000IU
Fresh egg mixture	1000.0mL
Glycerol	12.0mL

Source: This medium is available as a prepared medium from Oxoid.

Preparation of Medium: Add components to 1.0L of fresh egg mixture. Mix thoroughly. Gently heat and bring to boiling. Bring volume to 1640.0mL with distilled/deionized water. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C with tubes in an upright position.

Use: For the cultivation and maintenance of *Mycobacterium tuberculosis*.

Acid Products Test Broth

Composition per liter:

Invert sugar	10.0g
Peptone	10.0g
Yeast extract	7.5g

pH 4.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Cool to 25°C. Adjust pH to 4.0 with 25% tartaric acid solution. Distribute into screw-capped flasks in 300.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of acid tolerant microorganisms from foods. For the sterility testing of canned foods.

Acid Tomato Broth

Composition per liter:

Glucose	10.0g
Peptone	10.0g
Yeast extract	5.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Tomato juice	250.0mL
L-Cysteine solution	0.5mL

L-Cysteine Solution:

Composition per 10.0mL:

L-Cysteine	0.1g

Preparation of L-Cysteine Solution: Add 0.1g of L-cysteine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteine solution, to distilled/deionized water and bring volume to 999.5mL. Mix thoroughly. Adjust pH to 4.8. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 0.5mL of sterile L-cysteine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of a variety of fungi.

Acidic Tomato Medium for *Leuconostoc***Composition per liter:**

Agar (if needed)	15.0g
Glucose	10.0g
Peptone	10.0g
Yeast extract	5.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Tomato juice	250.0mL

pH 4.8 ± 0.2 at 25°C

Preparation of Medium: Add solid components to 750.0mL of distilled/deionized water. Add tomato juice. Mix well and warm gently until dissolved. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Leuconostoc oenos* and other *Leuconostoc* species.

**Acidophilic *Bacillus*
stearothermophilus Agar**

Composition per liter:

Part A	400.0mL
Part B	600.0mL

pH 5.0 ± 0.2 at 25°C

Part A:**Composition per 400.0mL:**

Soluble starch	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	1.0g
CaCl ₂ ·2H ₂ O	0.5g
MnCl ₂ ·4H ₂ O	0.5g

Preparation of Part A: Add components to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 4.7. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Part B:**Composition per 600.0mL:**

Agar	20.0g

Preparation of Part B: Add agar to distilled/deionized water and bring volume to 600.0mL. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Aseptically combine solution A and solution B. Mix thoroughly. Adjust pH to 5.0. Pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Bacillus stearothermophilus* and other acidophilic *Bacillus* species.

**Acidophilic *Bacillus*
stearothermophilus Broth**

Composition per liter:

Soluble starch	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	1.0g
CaCl ₂ ·2H ₂ O	0.5g
MnCl ₂ ·4H ₂ O	0.5g

pH 5.0 ± 0.2 at 25°C

Preparation of Medium: Dissolve all components except agar in 1.0L of distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Adjust to pH 5.0. Autoclave for 15 min at 15 psi pressure–121°C. Precipitate will dissolve after cooling and mixing.

Use: For the cultivation and maintenance of *Bacillus stearothermophilus* and other acidophilic *Bacillus* species.

**Actidione® Agar
(Cycloheximide Agar)**

Composition per liter:

Glucose	50.0g
Agar	15.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂ ·2H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	22.0mg
Actidione (cycloheximide)	10.0mg
FeCl ₃	2.5mg

pH 5.5 ± 0.2 at 25°C

Source: Actidione Agar is available as a prepared medium from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration and detection of bacteria in specimens containing large numbers of yeasts and molds.

**AE Sporulation Medium, Modified
(BAM M5)**

Composition per 1079.2mL:

Polypeptone™	10.0g
Yeast extract	10.0g
Na ₂ HPO ₄	4.36g

Ammonium acetate	1.5g
KH ₂ PO ₄	0.25g
MgSO ₄ ·7H ₂ O	0.2g
Raffinose solution	39.6mL
Na ₂ CO ₃ solution	13.2mL
CoCl ₂ ·6H ₂ O solution	13.2mL
Sodium ascorbate solution	13.2mL
pH 7.8 ± 0.1 at 25°C	

Raffinose Solution:**Composition per 100.0mL:**

Raffinose	10.0g

Preparation of Raffinose Solution: Add raffinose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Na₂CO₃ Solution:**Composition per 100.0mL:**

Na ₂ CO ₃	7.0g

Preparation of Na₂CO₃ Solution: Add Na₂CO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

CoCl₂ Solution:**Composition per 100.0mL:**

CoCl ₂ ·6H ₂ O	0.32g

Preparation of CoCl₂ Solution: Add CoCl₂·6H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Sodium Ascorbate Solution:**Composition per 100.0mL:**

Sodium ascorbate	1.5g

Preparation of Sodium Ascorbate Solution: Add sodium ascorbate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: Add components—except raffinose solution, Na₂CO₃ solution, CoCl₂ solution, and sodium ascorbate solution—to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.5 using 2m sodium carbonate solution. Distribute into tubes in 15.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 0.6mL of sterile raffinose solution, 0.2mL of sterile Na₂CO₃ solution, and 0.2mL of sterile CoCl₂ solution to each tube. Mix thoroughly. Prior to inoculation, steam medium for 10 min. Cool to 25°C. Aseptically add 0.2mL of sterile sodium ascorbate solution to each tube.

Use: For the cultivation and sporulation of *Clostridium perfringens*.

Aeromonas Differential Agar (Dextrin Fuchsin Sulfite Agar)

Composition per liter:

Dextrin	15.0g
Agar	13.0g
Pancreatic digest of casein	10.0g
Na ₂ HPO ₄	7.75g
NaCl	5.0g
Beef extract	3.0g
Na ₂ SO ₃	1.6g
Acid Fuchsin solution	50.0mL
pH 7.5 ± 0.2 at 25°C	

Acid Fuchsin Solution:**Composition per 50.0mL:**

Acid Fuchsin	0.25g

Preparation of Acid Fuchsin Solution: Add Acid Fuchsin to 50.0mL of 5% aqueous dioxan. Mix well to dissolve.

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contamination of the skin.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and differentiation of *Aeromonas* species from other Gram-negative rods such as *Pseudomonas* and Enterobacteriaceae. Specimens with low numbers of *Aeromonas* may first be enriched by growth in starch broth for 4–9 days. After 24 hrs of growth on this agar, colonies are sprayed with Nadi reagent (1% solution of *N,N,N',N'*-tetramethyl-*p*-phenylene-diammonium dichloride). A positive Nadi reaction (dextrin degradation) is indicated by a purple color at the periphery of the colony. Dextrin fermentation is also indicated by red colonies. *Aeromonas* species appear as large, convex, dark red colonies with a purple periphery.

Aeromonas hydrophila Medium

Composition per liter:

Inositol	10.0g
Pancreatic digest of casein	10.0g
L-Ornithine·HCl	5.0g
Proteose peptone	5.0g
Agar	3.0g
Yeast extract	3.0g
Mannitol	1.0g
Ferric ammonium citrate	0.5g

Na ₂ S ₂ O ₃ ·5H ₂ O	0.4g
Bromcresol Purple	0.02g
pH 6.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 6.7. Distribute into tubes in 5.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Aeromonas hydrophila*.

Aeromonas Medium **(Ryan's *Aeromonas Medium*)**

Composition per liter:

Agar	12.5g
Na ₂ S ₂ O ₃	10.67g
Proteose peptone	5.0g
NaCl	5.0g
Xylose	3.75g
L-Lysine·HCl	3.5g
Yeast extract	3.0g
Sorbitol	3.0g
Bile salts No. 3	3.0g
Inositol	2.5g
L-Arginine·HCl	2.0g
Lactose	1.5g
Ferric ammonium citrate	0.8g
Bromthymol Blue	0.04g
Thymol Blue	0.04g
Ampicillin	5.0mg
pH 8.0 ± 0.1 at 25°C	

Source: This medium is available as a dehydrated powder from Oxoid.

Preparation of Medium: Add components, except ampicillin, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 50°C and aseptically add 5.0mg of ampicillin. Pour into sterile Petri dishes.

Use: For the isolation and selective differentiation of *Aeromonas hydrophila* and other *Aeromonas* species. *Aeromonas* species appear as small (0.5–1.5mm), dark green colonies with darker centers. The addition of ampicillin improves the recovery of *Aeromonas* spp. *Plesiomonas* spp. as well as members of the Enterobacteriaceae also grow on this medium.

AFPA

(*Aspergillus flavus/parasiticus* Agar)

Composition per liter:

Yeast extract	20.0g
Agar	15.0g

Peptone	10.0g
Ferric ammonium citrate	0.5g
Dichloran (Botran®)	2.0mg
pH 6.3 ± 0.2 at 25°C	

Source: This medium is available as a dehydrated powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Add 100.0mg of chloramphenicol. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the selective isolation and enumeration of *Aspergillus flavus* and *Aspergillus parasiticus*. Colonies of these fungi appear with dark yellow-orange color on the reverse side.

Agar Medium for Differential Enumeration of Lactic Streptococci

Composition per 1170.0mL:

Agar	15.0g
Carboxymethylcellulose	15.0g
Calcium citrate	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
L-Arginine·HCl	5.0g
Casamino acids	2.5g
K ₂ HPO ₄	1.25g
Calcium carbonate solution	100.0mL
Nonfat milk solution	50.0mL
Bromcresol Purple solution	20.0mL
pH 5.9 ± 0.2 at 25°C	

Calcium Carbonate Solution:

Composition per 100.0mL:

CaCO ₃	3.0g

Preparation of Calcium Carbonate Solution:

Add CaCO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Nonfat Milk Solution:

Composition per 100.0mL:

Nonfat milk	11.0g

Preparation of Nonfat Milk Solution:

Add nonfat milk to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Bromcresol Purple Solution:

Composition per 20.0mL:

Bromcresol Purple	0.02g

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to distilled/deionized water

and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add agar to 500.0mL of distilled/deionized water. Gently heat and bring to boiling. In a separate flask, add carboxymethylcellulose and calcium citrate to 500.0mL of distilled/deionized water. Gently heat while stirring until a white, turbid suspension is formed. Combine the two solutions. Add the pancreatic digest of casein, yeast extract, K_2HPO_4 , casamino acids, and arginine. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 5.6 with 6N HCl. Distribute into screw-capped bottles in 100.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Immediately prior to pouring plates, aseptically add 5.0mL of sterile nonfat milk solution, 10.0mL of sterile calcium carbonate solution, and 2.0mL of sterile Bromocresol Purple solution to each screw-capped bottle. Mix thoroughly. The pH should be 5.9. Pour into cold, sterile Petri dishes.

Use: For the cultivation, differentiation, and enumeration of *Lactobacillus lactis*, *Lactobacillus lactis* subspecies *cremoris*, and *Lactobacillus lactis* subspecies *diacetylactis*. Lactose-fermenting bacteria such as *Lactobacillus lactis* subspecies *cremoris* appear as yellow colonies. Arginine-utilizing bacteria such as *Lactobacillus lactis* and *Lactobacillus lactis* subspecies *diacetylactis* appear as purple colonies. Citrate-utilizing bacteria such as *Lactobacillus lactis* subspecies *diacetylactis* appear as colonies surrounded by a clear zone.

Agar Medium P (PM Indicator Agar) (BAM M6)

Composition per liter:

Agar	15.0g
Glucose	5.25g
Peptone.....	5.0g
Beef extract.....	3.0g
Pancreatic digest of casein.....	1.7g
Tween 80.....	1.0g
NaCl.....	0.5g
Papaic digest of soybean meal.....	0.3g
K_2HPO_4	0.25g
Bromocresol Purple	0.06g
pH 7.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Bacillus stearothermophilus* for the detection of penicillin in milk.

AK Agar No. 2 (Sporulating Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	6.0g
Pancreatic digest of casein.....	4.0g
Yeast extract.....	3.0g
Beef extract.....	1.5g
Glucose	1.0g
$MnSO_4 \cdot 7H_2O$	0.3g
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C. Make sure medium is dissolved before autoclaving.

Use: For the preparation of spore suspensions used to detect antibiotic residues in milk and dairy products.

AKI Medium (BAM M7)

Composition per liter:

Peptone	15.0g
NaCl.....	5.0g
Yeast extract.....	4.0g
Sodium bicarbonate solution	30.0mL
pH 7.2 ± 0.2 at 25°C	

Sodium Bicarbonate Solution:

Composition per 100.0mL:

$NaHCO_3$	10.0g

Preparation of Sodium Bicarbonate Solution: Add sodium bicarbonate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: Add components, except sodium bicarbonate solution, to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile sodium bicarbonate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks. Prepare medium freshly.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

Alcal Mannose Medium**Composition per liter:**

K ₂ HPO ₄	15.1g
KH ₂ PO ₄	5.6g
Mannose.....	1.0g
Yeast extract.....	1.0g
Casamino acids.....	0.5g
MgSO ₄ ·7H ₂ O.....	0.4g
CaCl ₂ ·2H ₂ O.....	50.0mg
FeSO ₄ ·7H ₂ O.....	10.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus circulans*.

Alcaligenes Agar**Composition per liter:**

Agar.....	10.0g
Peptone.....	5.0g
Ammonium lactate.....	3.0g
Meat extract.....	3.0g
Ferric citrate.....	0.2g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add ferric citrate to distilled/deionized water and bring volume to 100.0mL. In a separate flask, add remaining components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.0. Steam the two solutions for 20 min on three consecutive days. Aseptically combine the two solutions. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Alcaligenes* species.

Alcaligenes Medium**Composition per liter:**

Peptone.....	5.0g
Beef extract.....	3.0g
Ferric citrate.....	0.2g
Ammonium lactate solution.....	3.0mL

pH 7.0 ± 0.2 at 25°C

Ammonium Lactate Solution:**Composition per 100.0mL:**

Lactic acid.....	60.0g

Preparation of Ammonium Lactate Solution: Dissolve lactic acid in 100.0mL of distilled/deionized water. Neutralize with NH₄OH to pH 7.0.

Preparation of Medium: Add peptone, beef extract, and ammonium lactate to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15

min at 15 psi pressure–121°C. Add ferric citrate aseptically. Mix thoroughly. Aseptically distribute into tubes or flasks.

Use: For the cultivation of *Alcaligenes tolerans*.

Alcaligenes Medium**Composition per liter:**

Tris.....	6.06g
NaCl.....	4.68g
KCl.....	1.49g
NH ₄ Cl.....	1.07g
Na ₂ SO ₄	0.43g
Na ₂ HPO ₄ ·12H ₂ O.....	0.23g
MgCl ₂ ·6H ₂ O.....	0.2g
CaCl ₂ ·2H ₂ O.....	0.03g
Ferric ammonium citrate.....	0.005g
Trace elements solution SL-7.....	1.0mL
Sodium succinate solution.....	10.0mL
CuSO ₄ solution.....	2.5mL

Sodium Succinate Solution:**Composition per 100.0mL:**

Sodium succinate.....	40.0g

Preparation of Sodium Succinate Solution:

Add sodium succinate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

CuSO₄ Solution:**Composition per 100.0mL:**

CuSO ₄	16.0g

Preparation of CuSO₄ Solution:

Add CuSO₄ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Trace Elements Solution SL-7:**Composition per 1001.0mL:**

CoCl ₂ ·6H ₂ O.....	200.0mg
MnCl ₂ ·4H ₂ O.....	100.0mg
ZnCl ₂	70.0mg
H ₃ BO ₃	60.0mg
Na ₂ MoO ₄ ·2H ₂ O.....	40.0mg
CuCl ₂ ·2H ₂ O.....	20.0mg
NiCl ₂ ·6H ₂ O.....	20.0mg
HCl (25%).....	1.0mL

Preparation of Trace Elements Solution SL-7:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add components, except CuSO₄ solution and sodium succinate solution, to distilled/deionized water and bring volume to 987.5mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile CuSO₄ solution and 2.5mL of sterile sodium

succinate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Alcaligenes* species.

***Alcaligenes* N5 Medium**

Composition per liter:

Sodium succinate·2H ₂ O	5.0g
KH ₂ PO ₄	0.75g
NH ₄ Cl	0.67g
K ₂ HPO ₄	0.61g
MgSO ₄ ·7H ₂ O	0.2g
CaCl ₂ ·2H ₂ O	0.03g
MnCl ₂ ·4H ₂ O	3.0mg
FeCl ₃	2.4mg
Na ₂ MoO ₄ ·2H ₂ O	1.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Alcaligenes faecalis*.

***Alcaligenes* NA YE Medium (*Alcaligenes* Nutrient Agar Yeast Extract Medium)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Yeast extract	5.0g
Beef extract	3.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Alcaligenes* species.

***Alcaligenes* NB YE Agar (*Alcaligenes* Nutrient Broth Yeast Extract Agar)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Yeast extract	5.0g
Beef extract	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Alcaligenes faecalis*.

***Alcaligenes* NB YE Broth (*Alcaligenes* Nutrient Broth Yeast Extract Broth)**

Composition per liter:

Pancreatic digest of gelatin	5.0g
Yeast extract	5.0g
Beef extract	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Alcaligenes faecalis*.

***Alcaligenes* NB YE Medium (*Alcaligenes* Nutrient Broth Yeast Extract Medium)**

Composition per liter:

Pancreatic digest of gelatin	5.0g
Yeast extract	5.0g
Beef extract	3.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Alcaligenes* species.

Alginate Utilization Medium

Composition per liter:

Solution B	500.0mL
Solution A	400.0mL
Solution C	100.0mL

Solution A:

Composition per 400.0mL:

Marine salts	38.0g

Preparation of Solution A: Add marine salts to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution B:**Composition** per 500.0mL:

Agar	20.0g
Sodium alginate	10.0g

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution C:**Composition** per 100.0mL:

Tris-HCl buffer.....	0.067g
NaNO ₃	0.047g
Ferric EDTA.....	66.5mg
Sodium glycerophosphate.....	6.67mg
Thiamine-HCl	67.0μg
Vitamin B ₁₂	1.3μg
Biotin	0.67μg

Preparation of Solution C: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine solutions A, B, and C. For liquid medium, omit agar from solution B.

Use: For the cultivation of microorganisms that can utilize alginate as a carbon source. Growth on alginate (production of alginase) is a diagnostic test used in the differentiation of *Vibrio* species.

Alkaline Bacillus Medium**Composition** per liter:

Agar	15.0g
Peptone.....	10.0g
Glucose	10.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	1.0g
Na ₂ CO ₃ solution	100.0mL
pH 8.5–11.0 at 25°C	

Na₂CO₃ Solution:**Composition** per 100.0mL:

Na ₂ CO ₃	10.0g

Preparation of Na₂CO₃ Solution: Add Na₂CO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except Na₂CO₃ solution, to distilled/deionized water and bring volume to 900.0mL. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 10 psi pressure–115°C. Cool to 45°–50°C. Aseptically add sterile Na₂CO₃ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of alkalophilic microorganisms such as *Bacillus alcalophilus*, *Bacillus circulans*, and other *Bacillus* species.

**Alkaline Peptone Agar
(BAM M8)****Composition** per liter:

NaCl.....	20.0g
Agar	15.0g
Peptone	10.0g
pH 8.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 8.5. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

**Alkaline Peptone Salt Broth
(APS Broth)
(BAM M9)****Composition** per liter:

NaCl.....	30.0g
Peptone	10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.5. Distribute into tubes in 10.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species from foods.

**Alkaline Peptone Water
(BAM M10)****Composition** per liter:

NaCl.....	10.0g
Peptone	10.0g
pH 8.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.5. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the cultivation and transport of *Vibrio cholerae* and other *Vibrio* species from foods.

Alkaline Peptone Water**Composition per liter:**

Peptone.....	10.0g
NaCl.....	5.0g
pH 9.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 9.0. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of alkalophilic microorganisms, especially *Vibrio* species.

Alkaline Polypectate Agar**Composition per liter:**

Agar.....	16.0g
Na ₂ CO ₃	10.0g
Peptone.....	6.0g
Sodium polypectate.....	5.0g
Yeast extract.....	3.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
MnSO ₄	40.0mg
pH 10.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Bring pH to 10.0. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of pectinolytic bacteria.

Alkaline Starch Agar**Composition per liter:**

Starch.....	20.0g
Agar.....	16.0g
Na ₂ CO ₃	10.0g
Peptone.....	6.0g
Yeast extract.....	3.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
MnSO ₄	40.0mg
pH 9.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Bring pH to 9.7. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of alkaliniphilic starch-utilizing bacteria.

Alkaline Xylan Agar**Composition per liter:**

Agar.....	15.0g
Larchwood xylan.....	10.0g
Polypeptone.....	5.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	0.45g
MgSO ₄ ·7H ₂ O.....	0.2g
pH 10.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically adjust pH with 10% sterile Na₂S₂O₃ solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* species.

Alkalophile Medium**Composition per liter:**

Agar.....	15.0g
Peptone.....	5.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g
Sodium sesquicarbonate solution.....	15.0mL
pH 9.5 ± 0.2 at 25°C	

Sodium Sesquicarbonate Solution:**Composition per 100.0mL:**

Sodium sesquicarbonate.....	9.0g

Preparation of Sodium Sesquicarbonate Solution: Add sodium sesquicarbonate to distilled/deionized water and bring volume to 100.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sodium sesquicarbonate solution, to distilled/deionized water and bring volume to 985.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 15.0mL of filter-sterilized sodium sesquicarbonate solution to adjust pH to 9.5. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Bacillus alcalophilus*, *Bacillus circulans*, *Bacillus submarinus*, and other *Bacillus* species.

Alkvisco Medium**Composition per liter:**

Agar.....	15.0g
Beef extract.....	10.0g

Peptone.....	10.0g
NaCl.....	5.0g
Acrylonitrile.....	0.5g
KCN.....	10.0mg

pH 6.5-8.0 at 25°C

Caution: Cyanide is toxic.

Preparation of Medium: Add components, except acrylonitrile, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 10 min at 15 psi pressure–121°C. Add acrylonitrile to 20.0mL of distilled/deionized water and filter sterilize. Add aseptically to the sterile basal medium.

Caution: Acrylonitrile is a carcinogen; use appropriate precautions.

Use: For the cultivation and maintenance of *Bacillus subtilis* and *Corynebacterium* species.

Allantoin Agar

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄ ·12H ₂ O	9.0g
NaCl.....	5.0g
KH ₂ PO ₄	1.5g
Meat extract	1.0g
Yeast extract.....	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
MnCl ₂ ·4H ₂ O	20.0mg
CaCl ₂	1.2mg
Glucose-allantoin solution	100.0mL

Glucose-Allantoin Solution:

Composition per 100.0mL:

Glucose	5.0g
Allantoin	1.0g

Preparation of Glucose-Allantoin Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except glucose-allantoin solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile glucose-allantoin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Bacillus* species.

ALOA Medium (Agar *Listeria* Ottavani & Agosti) (BAM M10a)

Composition per liter:

Agar	18.0g
Peptone	18.0g
LiCl.....	10.0g
Yeast extract.....	10.0g
Tryptone.....	6.0g
NaCl.....	5.0g
Na ₂ HPO ₄	2.5g
Na-pyruvate	2.0g
Glucose	2.0g
Mg-glycerophosphate	1.0g
MgSO ₄	0.5g
5-Bromo4-chloro-indolyl- β-D-glucopyranoside	0.05g
Phosphatidylinositol solution.....	50.0mL
Nalidixic acid solution.....	5.0mL
Ceftazidime solution.....	5.0mL
Cycloheximide solution.....	5.0mL
Polymyxin B solution	5.0mL

pH 7.2 ± 0.2 at 25°C

Nalidixic Acid Solution:

Composition per 5.0mL:

Nalidixic acid.....	0.02g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Ceftazidime Solution:

Composition per 5.0mL:

Ceftazidime.....	0.02g

Preparation of Ceftazidime Solution: Add ceftazidime to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 5.0mL:

Cycloheximide.....	0.05g
Ethanol.....	2.5mL

Preparation of Cycloheximide Solution: Add cycloheximide to 2.5mL of ethanol. Mix thoroughly. Bring volume to 5.0mL with distilled/deionized water. Filter sterilize.

Polymyxin B Solution:

Composition per 5.0mL:

Polymyxin B	76700U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Phosphatidylinositol Solution:**Composition per 50.0mL:**

L- α -phosphatidylinositol 2.0g

Preparation of Phosphatidylinositol Solution:

Add L- α -phosphatidylinositol to cold distilled/deionized water and bring volume to 50.0mL. Stir for 30 min so a homogeneous suspension is obtained. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48–50°C.

Preparation of Medium: Add components, except phosphatidylinositol solution, nalidixic acid solution, cetazidime solution, cycloheximide solution, and polymyxin B solution, to distilled/deionized water and bring volume to 930.0mL. Mix thoroughly. Adjust the pH to 7.2. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL sterile phosphatidylinositol solution, 5.0mL sterile nalidixic acid solution, 5.0mL sterile cetazidime solution, 5.0mL sterile cycloheximide solution, and 5.0mL sterile polymyxin B solution. Mix thoroughly. Pour into the Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria* spp. according to ISO standard 11290.

ALOA Medium
(Agar *Listeria* Ottavani & Agosti)
(BAM M10a)

Composition per liter:

Agar	18.0g
Peptone.....	18.0g
LiCl	10.0g
Yeast extract.....	10.0g
Tryptone.....	6.0g
NaCl.....	5.0g
Na ₂ HPO ₄	2.5g
Na-pyruvate	2.0g
Glucose	2.0g
Mg-glycerophosphate	1.0g
MgSO ₄	0.5g
5-Bromo4-chloro-indolyl- β -D-glucopyranoside	0.05g
Phosphatidylinositol solution.....	50.0mL
Amphotericin B solution.....	10.0mL
Nalidixic acid solution	5.0mL
Ceftazidime solution	5.0mL
Polymyxin B solution	5.0mL

pH 7.2 $\pm$ 0.2 at 25°C

Nalidixic Acid Solution:**Composition per 5.0mL:**

Nalidixic acid..... 0.02g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Ceftazidime Solution:**Composition per 5.0mL:**

Ceftazidime..... 0.02g

Preparation of Ceftazidime Solution: Add ceftazidime to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Amphotericin B Solution:**Composition per 10.0mL:**

Amphotericin B	0.01g
Dimethylformamide.....	7.5mL
HCL, 1M.....	2.5mL

Preparation of Amphotericin B Solution:

Add amphotericin B to 2.5mL of 1M HCL. Mix thoroughly. Add 7.5mL of dimethylformamide. Mix thoroughly. Filter sterilize.

Polymyxin B Solution:**Composition per 5.0mL:**

Polymyxin B 76,700U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Phosphatidylinositol Solution:**Composition per 50.0mL:**

L- α -phosphatidylinositol 2.0g

Preparation of Phosphatidylinositol Solution:

Add L- α -phosphatidylinositol to cold distilled/deionized water and bring volume to 50.0mL. Stir for 30 min so a homogeneous suspension is obtained. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48–50°C.

Preparation of Medium: Add components, except phosphatidylinositol solution, nalidixic acid solution, cetazidime solution, amphotericin B solution, and polymyxin B solution, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Adjust the pH to 7.2. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL sterile phosphatidylinositol solution, 5.0mL sterile nalidixic acid solution, 5.0mL sterile cetazidime solution, 10.0mL sterile amphotericin B solution, and 5.0mL sterile polymyxin B solution. Mix thoroughly. Pour into the Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria* spp. according to ISO standard 11290.

Amino Acid Assay Medium**Composition per liter:**

Glucose	50.0g
Sodium acetate	40.0g
NH ₄ Cl	6.0g
KH ₂ PO ₄	1.2g
K ₂ HPO ₄	1.2g
Asparagine	0.8g
L-Glutamic acid	0.6g
Pyridoxamine-HCl	0.6g
Pyridoxal-HCl	0.6g
DL-Valine	0.5g
L-Lysine-HCl	0.5g
DL-Isoleucine	0.5g
DL-Leucine	0.5g
L-Arginine-HCl	0.5g
DL-Threonine	0.4g
MgSO ₄ ·7H ₂ O	0.4g
DL-Alanine	0.4g
DL-Phenylalanine	0.2g
L-Tyrosine	0.2g
Lysine	0.2g
L-Aspartic acid	0.2g
DL-Methionine	0.2g
L-Proline	0.2g
L-Histidine-HCl	0.12g
DL-Serine	0.1g
L-Cystine	0.1g
DL-Tryptophan	0.08g
MnSO ₄ ·7H ₂ O	0.04g
FeSO ₄	0.02g
NaCl	0.02g
Adenine sulfate	0.02g
Guanine-HCl	0.02g
Uracil	0.02g
Xanthine	0.02g
Nicotinic acid	2.0mg
Pyridoxine-HCl	2.0mg
Thiamine-HCl	1.0mg
Calcium pantothenate	1.0mg
Riboflavin	1.0mg
<i>p</i> -Aminobenzoic acid	0.2mg
Folic acid	0.02mg
Biotin	2.0μg

pH 6.7 ± 0.2 at 25°C

Source: This medium is available, without cystine, lysine or methionine, as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to 1.0L of distilled water, omitting the specific amino acid to be assayed for in the procedure. Heat to boiling for 2–3 min. Distribute into tubes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the microbiological assay for amino acids. *Pediococcus acidilactici* ATCC 8042 and *Enterococcus hirae* ATCC 8043 are used as test microorganisms.

**Anaerobe Agar
(BAM M11)****Composition per 1001.5mL:**

Agar	15.0g
Pancreatic digest of casein	15.0g
Pancreatic digest of soybean meal	5.0g
NaCl	5.0g
L-Cysteine-HCl·H ₂ O solution	5.0mL
Vitamin K ₁ solution	1.0mL
Hemin solution	0.5mL

pH 7.0 ± 0.2 at 25°C

L-Cysteine-HCl·H₂O Solution:**Composition per 5.0mL:**

L-Cysteine-HCl·H ₂ O	0.4g
NaOH (1 <i>N</i> solution)	5.0mL

Preparation of L-Cysteine-HCl·H₂O Solution: Add L-cysteine-HCl·H₂O to 5.0mL 1*N* NaOH. Mix thoroughly. Filter sterilize.

Hemin Solution:**Composition per 100.0mL:**

Hemin	1.0g

Preparation of Hemin Solution: Add hemin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool. Refrigerate at 4°C for storage.

Vitamin K₁ Solution:**Composition per 100.0mL:**

Vitamin K ₁	1.0g
Ethanol, absolute	20.0mL

Preparation of Vitamin K₁ Solution: Add vitamin K₁ to 100.0mL of 95% ethanol. Mix thoroughly. Solution may require 2–3 days with intermittent shaking to completely dissolve. Filter sterilize. Refrigerate at 4°C for storage.

Preparation of Medium: Add components, ex-hemin solution and vitamin K₁ solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0 ± 0.2 at 25°C. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.5mL of sterile hemin solution and 1.0mL of sterile vitamin K₁ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Reduce medium for 24 hr by incubation in an anaerobic glove box of GasPak jar prior to use.

Use: For the cultivation of anaerobic bacteria, such as *Brucella* and *Clostridium* spp.

Anaerobe Agar (LMG Medium 41)

Composition per liter:

Agar Base.....	800.0mL
Solution A.....	100.0mL
Solution B.....	100.0mL
pH 6.9 ± 0.2 at 25°C	

Agar Base:

Composition 800.0mL:

Agar.....	30.5g
Tryptone.....	5.0g
Yeast extract.....	5.0g
(NH ₄) ₂ SO ₄	0.5g
MgSO ₄ ·7H ₂ O.....	0.1g
Sodium thioglycolate.....	0.5g
Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O.....	55.0mg
Na ₂ MoO ₄ ·2H ₂ O.....	2.4mg
Na ₂ SeO ₃ ·5H ₂ O.....	0.23mg

Preparation of Agar Base: Add components to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Solution A:

Composition 100.0mL:

Glucose.....	18.0g
K ₂ HPO ₄	7.0g
KH ₂ PO ₄	5.5g

Preparation of Solution A: Add components to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Solution B:

Composition 100.0mL:

NaHCO ₃	10.0g

Preparation of Solution B: Add components to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically add solutions A and B to the agar base. Adjust pH to 6.9. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Incubate anaerobically under 100% CO₂ gas atmosphere.

Use: For the cultivation of anerobic bacteria.

Anaerobe Medium (LMG Medium 41)

Composition per liter:

Agar Base.....	800.0mL

Solution A.....	100.0mL
Solution B.....	100.0mL
pH 6.9 ± 0.2 at 25°C	

Agar Base:

Composition 800.0mL:

Tryptone.....	5.0g
Yeast extract.....	5.0g
(NH ₄) ₂ SO ₄	0.5g
MgSO ₄ ·7H ₂ O.....	0.1g
Sodium thioglycolate.....	0.5g
Agar.....	0.5g
Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O.....	55.0mg
Na ₂ MoO ₄ ·2H ₂ O.....	2.4mg
Na ₂ SeO ₃ ·5H ₂ O.....	0.23mg

Preparation of Agar Base: Add components to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Solution A:

Composition 100.0mL:

Glucose.....	18.0g
K ₂ HPO ₄	7.0g
KH ₂ PO ₄	5.5g

Preparation of Solution A: Add components to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Solution B:

Composition 100.0mL:

NaHCO ₃	10.0g

Preparation of Solution B: Add components to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically add solutions A and B to the sterila Agar Base. Adjust pH to 6.9. Mix thoroughly. Distribute into sterile tubes. Incubate anaerobically under 100% CO₂ gas atmosphere.

Use: For the cultivation of anerobic bacteria.

Anaerobic Agar

Composition per liter:

Agar.....	20.0g
Pancreatic digest of casein.....	20.0g
Glucose.....	10.0g
NaCl.....	5.0g
Sodium thioglycolate.....	2.0g
Sodium formaldehyde sulfoxylate.....	1.0g
Methylene Blue.....	2.0mg
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.2. Distribute into tubes until medium is 3 inches deep. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of anaerobic microorganisms, especially *Clostridium* species.

Anaerobic Agar

Composition per liter:

Pancreatic digest of casein	17.5g
Agar	15.0g
Glucose	10.0g
Papaic digest of soybean meal	2.5g
NaCl	2.5g
Sodium thioglycolate	2.0g
Sodium formaldehyde sulfoxylate	1.0g
L-Cystine	0.4g
Methylene Blue	2.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Use with Brewer anaerobic Petri dishes or in tubes or ordinary plates and incubate in anaerobic jars.

Use: For the cultivation of *Clostridium* species and for anaerobic microorganisms.

Anaerobic Broth

Composition per liter:

Pancreatic digest of casein	17.5g
Glucose	10.0g
NaCl	2.5g
Papaic digest of soybean meal	2.5g
Sodium thioglycolate	2.0g
Sodium formaldehyde sulfoxylate	1.0g
L-Cystine	0.4g
Methylene Blue	2.0mg

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of anaerobic and microaerophilic microorganisms.

Anaerobic CNA Agar (Anaerobic Colistin Nalidixic Acid Agar)

Composition per liter:

Agar	13.0g
Pancreatic digest of casein	12.0g
NaCl	5.0g
Peptic digest of animal tissue	5.0g
Yeast extract	3.0g
Beef extract	3.0g
Cornstarch	1.0g
Glucose	1.0g
L-Cysteine-HCl·H ₂ O	0.5g
Vitamin K ₁	10.0mg
Hemin	10.0mg
Colistin	10.0mg
Nalidixic acid	10.0mg
Sheep blood, defibrinated	50.0mL

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of anaerobic streptococci.

Anaerobic Egg Yolk Agar

Composition per 1080.0mL:

Agar	20.0g
Proteose peptone	20.0g
NaCl	5.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
Egg yolk emulsion, 50%	80.0mL

pH 7.0 ± 0.2 at 25°C

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack 11 eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components, except egg yolk emulsion, 50%, to distilled/deionized

water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 80.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Allow plates to dry at 35°C for 24 hr.

Use: For the cultivation of *Clostridium* species.

Anaerobic Egg Yolk Agar

Composition per liter:

Agar	20.0g
Proteose peptone	20.0g
Pancreatic digest of casein	5.0g
NaCl	5.0g
Yeast extract	5.0g
Egg yolk emulsion, 50%	20.0mL
pH 7.0 ± 0.2 at 25°C	

Egg Yolk Emulsion, 50%:

Composition per 20.0mL:

Chicken egg yolks	2
NaCl (0.9% solution)	10.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Measure 10.0mL of egg yolk emulsion and add to 10.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry at 35°C for 24 hr.

Use: For the cultivation of *Yersinia enterocolitica*.

**Anaerobic Egg Yolk Agar
(BAM M12)**

Composition per liter:

Agar	20.0g
Proteose peptone	20.0g
Pancreatic digest of casein	5.0g
NaCl	5.0g
Yeast extract	5.0g
Egg yolk emulsion, 50%	80.0mL
pH 7.0 ± 0.2 at 25°C	

Egg Yolk Emulsion, 50%:

Composition per 80.0mL:

Chicken egg yolks	2 or more
NaCl (0.85% solution)	40.0mL

Preparation of Egg Yolk Emulsion, 50%:

Wash fresh eggs with stiff brush and drain. Soak eggs in 70% ethanol for 1 hr. Crack eggs aseptically and separate yolks from whites. Drain contents of yolk sacs into sterile stoppered graduate cylinder and discard sacs. Measure 40.0mL of egg yolk emulsion and add 40.0mL of 0.85% NaCl solution. Mix thoroughly by inverting graduate cylinder. Warm to 45°–50°C.

Preparation of Medium:

Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 80.0mL sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry at ambient temperature for 2–3 days or at 35°C for 24 hr. Check plates for contamination before use.

Use: For the cultivation of *Yersinia enterocolitica*.

Anaerobic TVLS Medium

Composition per liter:

Pancreatic digest of casein	17.0g
Beef extract	7.5g
Glucose	6.0g
Enzymatic hydrolysate of soybean meal	3.0g
Liver hydrolysate	3.0g
NaCl	2.5g
Na ₂ SO ₃	0.7g
Sodium thioglycolate	0.5g
L-Cysteine-HCl·H ₂ O	0.25g
Agar	0.1g
Bovine serum	100.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except bovine serum, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of bovine serum. Distribute into sterile tubes.

Use: For the isolation and cultivation of anaerobic microorganisms.

Andersen's Pork Pea Agar

Composition per 1685.0mL:

Agar	16.0g
Peptone	5.0g
Pancreatic digest of casein	1.6g
K ₂ HPO ₄	1.25g
Soluble starch	1.0g
Sodium thioglycolate	0.5g
Pork infusion	800.0mL
Thioglycolate agar	660.0mL

Pea infusion.....	200.0mL
NaHCO ₃ solution	25.0mL
pH 7.2 ± 0.2 at 25°C	

Pork Infusion:**Composition** per liter:

Pork, fresh lean ground.....	454.0g

Preparation of Pork Infusion: Add ground pork to distilled/deionized water and bring volume to 1.0L. Autoclave for 60 min at 0 psi pressure–100°C. Filter through two layers of cheesecloth. Cool to 4°C. Skim fat from surface. Warm to 25°C. Centrifuge at 5000 rpm for 10 min. Discard pellet.

Pea Infusion:**Composition** per 450.0mL:

Green peas, fresh or frozen.....	454.0g
Diatomaceous earth (celite)	10.0g

Preparation of Pea Infusion: Add green peas to 450.0mL of distilled/deionized water. Blend until smooth. Autoclave for 60 min at 0 psi pressure–100°C. Centrifuge at 5000 rpm for 10 min. Discard pellet. Clarify supernatant solution with diatomaceous earth (celite). Filter through Whatman #4 filter paper. Use filtrate solution.

Thioglycolate Agar:**Composition** per liter:

Agar	20.75g
Pancreatic digest of casein.....	15.0g
Glucose	5.5g
Yeast extract.....	5.0g
NaCl.....	2.5g
L-Cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin	1.0mg

pH 7.1 ± 0.2 at 25°C

Preparation of Thioglycolate Agar: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

NaHCO₃ Solution:**Composition** per 100.0mL:

NaHCO ₃	5.0g

Preparation of NaHCO₃ Solution: Add NaHCO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Combine components, except NaHCO₃ solution and thioglycolate agar. Mix thoroughly. Adjust pH to 7.2. Autoclave for 5 min at 15 psi pressure–121°C. While medium is still hot, add 25.0g of celite. Filter through Whatman #4 filter paper with suction. Autoclave for 12 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add

25.0mL of sterile NaHCO₃ solution. Mix thoroughly. Pour into sterile Petri dishes in 15.0mL volumes. Allow agar to solidify. Cover agar with 10.0mL of sterile, cooled thioglycolate agar.

Use: For the cultivation of mesophilic *Clostridium* species. For the recovery of endospores from foods following heat treatments.

Andrade's Broth**Composition** per liter:

Pancreatic digest of gelatin.....	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Andrade's indicator.....	10.0mL
Carbohydrate solution.....	50.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a prepared medium from BD Diagnostic Systems, in tubes containing adonitol, arabinose, cellobiose, glucose, dulcitol, fructose, galactose, inositol, lactose, maltose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose, trehalose, or xylose.

Andrade's Indicator**Composition** per 100.0mL:

NaOH (1N solution).....	16.0mL
Acid Fuchsin.....	0.1g

Preparation of Andrade's Indicator: Add Acid Fuchsin to NaOH solution and bring volume to 100.0mL with distilled/deionized water.

Carbohydrate Solution:**Composition** per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Adonitol, arabinose, cellobiose, glucose, dulcitol, fructose, galactose, inositol, lactose, maltose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose, trehalose, xylose, or other carbohydrates may be used. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute in 10.0mL volumes into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Add 0.5mL of sterile carbohydrate solution to each tube.

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Use: For the determination of carbohydrate fermentation reactions of microorganisms, particularly mem-

bers of the Enterobacteriaceae. A Durham tube is used to collect gas produced during the fermentation reaction. Acid production is indicated by a pink color.

Andrade's Carbohydrate Broth and Indicator (BAM M13)

Composition per liter:

Pancreatic digest of gelatin	10.0g
NaCl	10.0g
Beef extract	3.0g
Carbohydrate solution	100.0mL
Andrade's indicator	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a prepared medium from BD Diagnostic Systems, in tubes containing adonitol, arabinose, cellobiose, glucose, dulcitol, fructose, galactose, inositol, lactose, maltose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose, trehalose, or xylose.

Andrade's Indicator

Composition per 26.0mL:

NaOH (1N solution)	16.0mL
Acid Fuchsin	0.21g

Preparation of Andrade's Indicator: Add Acid Fuchsin to NaOH solution and bring volume to 26.0mL with distilled/deionized water.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. For glucose, lactose, sucrose, and mannitol, add 10.0g to distilled/deionized water and bring volume to 100.0mL. For dulcitol, salicin, and other carbohydrates, add 5.0g to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool. Aseptically add 100mL of sterile carbohydrate solution to 900mL of sterile medium. Mix thoroughly. Aseptically distribute into tubes or flasks. Alternately, prior to autoclaving, distribute 9.0mL volumes into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Add 1.0mL of sterile carbohydrate solution to each tube.

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Use: For the determination of carbohydrate fermentation reactions of microorganisms, particularly members of the Enterobacteriaceae. A Durham tube is used to collect gas produced during the fermentation reaction. Acid production is indicated by a pink color.

Anthranilic Acid Medium, Revised

Composition per 1040.0mL:

Na ₂ HPO ₄	6.0g
KH ₂ PO ₄	3.0g
NH ₄ Cl	1.0g
NaCl	0.5g
Glucose solution	25.0mL
CaCl ₂ solution	10.0mL
MgSO ₄ solution	10.0mL
Anthranilic acid solution	5.0mL

Glucose Solution:

Composition per 100.0mL:

D-Glucose	20.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

CaCl₂ Solution:

Composition per 100.0mL:

CaCl ₂ ·2H ₂ O	0.147g

Preparation of CaCl₂ Solution: Add CaCl₂ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

MgSO₄ Solution:

Composition per 10.0mL:

MgSO ₄ ·7H ₂ O	2.47g

Preparation of MgSO₄ Solution: Add MgSO₄ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Anthranilic Acid Solution:

Composition per 100.0mL:

Anthranilic acid	1.0g
Ethanol (95% solution)	100.0mL

Preparation of Anthranilic Acid Solution: Add anthranilic acid to 100.0mL of ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, CaCl₂ solution, MgSO₄ solution, and anthranilic acid solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 25.0mL of sterile glucose solution, 10.0mL of sterile CaCl₂ solution, 10.0mL of sterile MgSO₄ solution, and 5.0mL of sterile an-

thranilic acid solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Escherichia coli*.

**Antibiotic Medium 1
(Penassay Seed Agar)
(Seed Agar)/(Agar Medium A)
(BAM M14)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Pancreatic digest of casein	4.0g
Yeast extract	3.0g
Beef extract	1.5g
Glucose	1.0g

pH 6.6 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For antibiotic assay testing, detection of antibiotics in milk, and determination of the antimicrobial effectiveness of antibiotics.

Antibiotic Medium 1 with Tetracycline

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Pancreatic digest of casein	4.0g
Yeast extract	3.0g
Beef extract	1.5g
Glucose	1.0g
Tetracycline solution	10.0mL

pH 6.6 ± 0.1 at 25°C

Tetracycline Solution:

Composition per 10.0mL:

Tetracycline	0.02g

Preparation of Tetracycline Solution: Add tetracycline to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except tetracycline solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile tetracycline solution. Mix thoroughly.

Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective cultivation and maintenance of *Salmonella choleraesuis*.

**Antibiotic Medium 1 with Tetracycline,
Streptomycin, and Chloramphenicol**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Pancreatic digest of casein	4.0g
Yeast extract	3.0g
Beef extract	1.5g
Glucose	1.0g
Antibiotic solution	10.0mL

pH 6.6 ± 0.1 at 25°C

Antibiotic Solution:

Composition per 10.0mL:

Tetracycline	0.02g
Streptomycin	0.02g
Chloramphenicol	0.02g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except tetracycline solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective cultivation and maintenance of *Salmonella choleraesuis*.

Antibiotic Medium 2

(Base Agar)

(Penassay Base Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Yeast extract	3.0g
Beef extract	1.5g

pH 6.6 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For use as a base layer in antibiotic assay testing. Especially useful for the plate assay of bacitracin and penicillin G.

Antibiotic Medium 3 (Penassay Broth)

Composition per liter:

Pancreatic digest of gelatin.....	5.0g
NaCl.....	3.5g
Yeast extract.....	1.5g
Beef extract.....	1.5g
Glucose.....	1.0g
K ₂ HPO ₄	3.68g
KH ₂ PO ₄	1.32g

pH 7.0 ± 0.05 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For antibiotic assay testing. Used for the serial dilution assay of penicillins and other antibiotics. Used in the turbidimetric assay of penicillin and tetracycline with *Staphylococcus aureus*. For the cultivation and maintenance of *Bacillus subtilis*, *Salmonella choleraesuis*, and *Staphylococcus aureus*. For the cloning of plasmids in *Streptococcus mutans*.

Antibiotic Medium 3 Plus

Composition per liter:

Agar.....	15.0g
Peptone.....	5.0g
K ₂ HPO ₄	3.68g
NaCl.....	3.5g
Yeast extract.....	2.5g
Glucose.....	1.75g
Beef extract.....	1.5g
KH ₂ PO ₄	1.32g

pH 7.0 ± 0.05 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For antibiotic assay testing and for the cultivation of *Escherichia coli*.

Antibiotic Medium 4 (Yeast Beef Agar) (Agar Medium B) (BAM M15)

Composition per liter:

Agar.....	15.0g
Pancreatic digest of gelatin.....	6.0g
Yeast extract.....	3.0g
Beef extract.....	1.5g
Glucose.....	1.0g

pH 6.6 ± 0.05 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For antibiotic assay testing.

Antibiotic Medium 5 (Streptomycin Assay Agar with Yeast Extract)

Composition per liter:

Agar.....	15.0g
Pancreatic digest of gelatin.....	6.0g
Yeast extract.....	3.0g
Beef extract.....	1.5g

pH 7.9 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing. For the streptomycin assay using the cylinder plate technique and *Bacillus subtilis* as test organism.

Antibiotic Medium 6

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
Glucose.....	2.5g

K ₂ HPO ₄	2.5g
MnSO ₄ ·H ₂ O	0.03g
pH 7.0 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing.

Antibiotic Medium 7

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Yeast extract	3.0g
Beef extract	1.5g
pH 7.0 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For use as a base layer in antibiotic assay testing. Especially useful for the plate assay of bacitracin and penicillin G.

Antibiotic Medium 8 (Base Agar with Low pH)

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Yeast extract	3.0g
Beef extract	1.5g
pH 5.9 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing. For use as the base agar and the seed agar in the plate assay of tetracycline. For use as the seed agar in the plate assay of vancomycin, mitomycin, and mithramycin.

Antibiotic Medium 9 (Polymyxin Base Agar)

Composition per liter:

Agar	20.0g
Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g
pH 7.2 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing. For base agar for the plate assay of carbenicillin, colistimethate, and polymyxin B.

Antibiotic Medium 10 (Polymyxin Seed Agar)

Composition per liter:

Pancreatic digest of casein	17.0g
Agar	12.0g
Polysorbate 80	10.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing. For seed agar for the plate assay of carbenicillin, colistimethate, and polymyxin B.

Antibiotic Medium 11 (Neomycin Assay Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	6.0g
Pancreatic digest of casein	4.0g
Yeast extract	3.0g

Beef extract.....	1.5g
Glucose	1.0g
pH 8.0 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay testing. For base agar and seed agar for the plate assay to test the effectiveness of neomycin sulfate, amoxicillin, ampicillin, clindamycin, cyclacillin, erythromycin, gentamycin, neomycin, oleandomycin, and sisomycin.

Antibiotic Medium 12

Composition per liter:

Agar	25.0g
Peptone.....	10.0g
Glucose	10.0g
NaCl.....	10.0g
Yeast extract.....	5.0g
Beef extract.....	2.5g
pH 6.0 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For antibiotic assay effectiveness testing.

Antibiotic Medium 13 (Sabouraud Liquid Broth, Modified) (Fluid Sabouraud Medium)

Composition per liter:

Glucose	20.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
pH 5.7 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For testing the effectiveness of antibiotics on yeast and molds.

Antibiotic Sulfonamide Sensitivity Test Agar (ASS Agar)

Composition per liter:

Agar	12.0g
Proteose peptone.....	10.0g
Beef extract.....	10.0g
NaCl.....	3.0g
Glucose	2.0g
Na ₂ HPO ₄	2.0g
Sodium acetate.....	1.0g
Adenine.....	0.01g
Guanine.....	0.01g
Uracil	0.01g
Xanthine.....	0.01g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For testing the antimicrobial effectiveness of antibiotics and sulfonamides. For detecting the presence of antimicrobial substances in milk, urine, and other fluids.

Antibiotic Sulfonamide Sensitivity Test Agar (ASS Agar)

Composition per liter:

Agar	12.0g
Proteose peptone.....	10.0g
Beef extract.....	10.0g
NaCl.....	3.0g
Glucose	2.0g
Na ₂ HPO ₄	2.0g
Sodium acetate.....	1.0g
Adenine.....	0.01g
Guanine.....	0.01g
Uracil	0.01g
Xanthine.....	0.01g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For detecting the presence of antimicrobial substances in milk.

AOAC Letheen Broth (Association of Official Analytical Chemists Letheen Broth)

Composition per liter:

Peptic digest of animal tissue.....	10.0g
Polysorbate 80.....	5.0g
NaCl.....	5.0g
Beef extract.....	5.0g
Lecithin.....	0.7g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the determination of phenol coefficients of disinfectant products containing cationic surface-active materials. Use according to *Official Methods of Analysis of the Association of Official Analytical Chemists* (AOAC).

Aolpha Medium

Composition per 1041.0mL:

NaCl.....	100.0g
Agar.....	15.0g
MgSO ₄ ·7H ₂ O.....	9.5g
MgCl ₂ ·6H ₂ O.....	5.0g
KCl.....	5.0g
Peptone.....	5.0g
Yeast extract.....	1.0g
CaCl ₂ ·2H ₂ O.....	0.2g
(NH ₄) ₂ SO ₄	0.1g
KNO ₃	0.1g
Metals solution.....	20.0mL
Phosphate solution.....	20.0mL
Vitamin solution.....	1.0mL

pH 7.0 ± 0.2 at 25°C

Metals Solution:

Composition per liter:

MgSO ₄ ·7H ₂ O.....	29.7g
Nitrilotriacetic acid.....	10.0g
CaCl ₂ ·2H ₂ O.....	3.3g
FeSO ₄ ·7H ₂ O.....	99.0mg
Na ₂ MoO ₄ ·2H ₂ O.....	12.7mg
Metals “44”.....	50.0mL

Preparation of Metals Solution: Solubilize nitrilotriacetic acid with KOH. Dissolve remaining ingredients. Adjust pH to 7.2 with KOH or H₂SO₄. Autoclave for 15 min at 15 psi pressure–121°C. Add aseptically to sterile basal medium.

Metals “44”:

Composition per 100.0mL:

ZnSO ₄ ·7H ₂ O.....	1.1g
FeSO ₄ ·7H ₂ O.....	0.5g
EDTA.....	0.25g
MnSO ₄ ·7H ₂ O.....	0.154g
CuSO ₄ ·5H ₂ O.....	0.04g
Co(NO ₃) ₂ ·6H ₂ O.....	0.025g
Na ₂ B ₄ O ₇ ·10H ₂ O.....	0.018g

Preparation of Metals “44”: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Add aseptically to sterile basal medium.

Phosphate Solution:

Composition per liter:

K ₂ HPO ₄	2.5g
KH ₂ PO ₄	2.5g

Preparation of Phosphate Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Add aseptically to sterile basal medium.

Vitamin Solution:

Composition per liter:

Pyridoxine-HCl.....	10.0mg
Calcium pantothenate.....	5.0mg
Nicotinamide.....	5.0mg
Riboflavin.....	5.0mg
Thiamine-HCl.....	5.0mg
Biotin.....	2.0mg
Folic acid.....	2.0mg
Cyanocobalamin.....	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize and add aseptically to sterile basal medium.

Preparation of Medium: Add components—except Metals “44”, phosphate solution, and vitamin solution—to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH of basal medium to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C and aseptically add the Metals “44”, phosphate, and vitamin solutions.

Use: For the cultivation and maintenance of *Halomonas meridiana* and other *Halomonas* species.

Apple Juice Yeast Extract Medium (AJYE Medium)

Composition per 1200.0mL:

Agar	30.0g
Yeast extract	10.0g
Apple juice	1.0L
pH 4.8 ± 0.2 at 25°C	

Preparation of Medium: Add yeast extract to 1.0L of apple juice. Mix thoroughly. Adjust pH to 4.8. Autoclave for 10 min at 9 psi pressure—114°C. Cool to 45°–50°C. In a separate flask, add agar to 200.0mL of distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 45°–50°C. Aseptically combine the sterile apple juice solution with the sterile agar solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of *Zymomonas* species.

APT Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	12.5g
Glucose	10.0g
Yeast extract	7.5g
NaCl	5.0g
K ₂ HPO ₄	5.0g
Sodium citrate	5.0g
Na ₂ CO ₃	1.25g
MnCl ₂ ·4H ₂ O	0.14g
MgSO ₄ ·7H ₂ O	0.8g
Polysorbate 80	0.2g
FeSO ₄ ·7H ₂ O	0.04g
Thiamine·HCl	1.0mg
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi—118°–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of bacteria, especially heterofermentative lactobacilli, from meat and other foods. For the cultivation of streptococci.

APT Agar

Composition per liter:

Agar	13.5g
Pancreatic digest of casein	10.0g
Glucose	10.0g
Yeast extract	7.5g

NaCl	5.0g
KH ₂ PO ₄	5.0g
Sodium citrate	5.0g
Na ₂ CO ₃	1.25g
MnCl ₂ ·4H ₂ O	0.14g
MgSO ₄ ·7H ₂ O	0.8g
Polysorbate 80	0.2g
FeSO ₄ ·7H ₂ O	0.04g

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi—118°–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of bacteria, especially heterofermentative lactobacilli, from meat and other foods. For the cultivation of streptococci.

APT Broth

Composition per liter:

Pancreatic digest of casein	12.5g
Glucose	10.0g
Yeast extract	7.5g
NaCl	5.0g
K ₂ HPO ₄	5.0g
Sodium citrate	5.0g
MgSO ₄ ·7H ₂ O	0.8g
MnCl ₂ ·4H ₂ O	0.14g
FeSO ₄ ·7H ₂ O	0.04g
Polysorbate 80	0.2g
Na ₂ CO ₃	1.25g
Thiamine·HCl	1.0mg
pH 7.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi—118°–121°C.

Use: For the cultivation of lactic acid bacteria. For the cultivation of heterofermentative lactobacilli from meat and other foods. The American Public Health Association recommends adding 100.0µg/L of thiamine to this medium.

APT Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Glucose	10.0g

Yeast extract.....	7.5g
NaCl.....	5.0g
KH ₂ PO ₄	5.0g
Sodium citrate.....	5.0g
Na ₂ CO ₃	1.25g
MgSO ₄ ·7H ₂ O.....	0.8g
MnCl ₂ ·4H ₂ O.....	0.14g
FeSO ₄ ·7H ₂ O.....	0.04g
Polysorbate 80.....	0.2g
pH 7.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi—118°–121°C.

Use: For the cultivation of lactic acid bacteria. For the cultivation of heterofermentative lactobacilli from meat and other foods. The American Public Health Association recommends adding 100.0µg/L of thiamine to this medium.

Arginine Broth (BAM M44)

Composition per liter:

L-Arginine.....	5.0g
Peptone or gelysate.....	5.0g
Yeast extract.....	3.0g
Glucose.....	1.0g
Bromcresol Purple.....	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure—121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate the amino acid arginine. Bacteria that decarboxylate arginine turn the medium turbid purple.

Arginine Broth with NaCl (BAM M44)

Composition per liter:

L-Arginine.....	5.0g
Peptone or gelysate.....	5.0g
Yeast extract.....	3.0g

Glucose.....	1.0g
Bromcresol Purple.....	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure—121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of *Vibrio* spp. based on their ability to decarboxylate the amino acid arginine. Bacteria that decarboxylate arginine turn the medium turbid purple.

Arginine Glucose Slants (AGS) (BAM M16)

Composition per liter:

NaCl.....	20.0g
Agar.....	13.5g
Pancreatic digest of casein.....	10.0g
L-Arginine-HCl.....	5.0g
Peptone.....	5.0g
Yeast extract.....	3.0g
Glucose.....	1.0g
Ferric ammonium citrate.....	0.5g
Na ₂ S ₂ O ₃ ·5H ₂ O.....	0.3g
Bromcresol Purple.....	0.02g
pH 6.8–7.0 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 12 min at 15 psi pressure—121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of *Vibrio* species.

Armstrong *Fusarium* Medium

Composition per liter:

Glucose.....	20.0g
Ca(NO ₃) ₂ ·4H ₂ O.....	8.4g
KH ₂ PO ₄	1.09g
KCl.....	0.22g
FeCl ₃	0.2µg
MnSO ₄	0.2µg
ZnSO ₄	0.2µg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: For the cultivation of *Fusarium* species.

Ascospore Agar**Composition per liter :**

Potassium acetate	30.0g
Yeast extract	2.5g
Glucose	1.0g

pH 6.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enrichment of ascosporegenous yeasts and their production of ascospores.

ASLA Agar**Composition per liter:**

Sodium lactate	20.0g
Davis agar	10.0g
(NH ₄) ₂ SO ₄	3.0g
Na ₂ HPO ₄	1.2g
L-Cysteine·HCl	0.5g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
FeSO ₄ ·7H ₂ O	0.04g
Vitamin solution	10.0mL

pH 6.5 ± 0.2 at 25°C

Vitamin Solution:**Composition per liter:**

Biotin	0.1g
Calcium pantothenate	0.1g
<i>p</i> -Aminobenzoic acid	0.1g
Thiamine	0.1g

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Store solution at –20°C.

Preparation of Medium: Add components, except vitamin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile vitamin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of most *Propionibacterium* species from foods.

Asparaginate Glycerol Agar**Composition per liter:**

Agar	15.0g
Sodium asparaginate	1.0g

K ₂ HPO ₄	1.0g
Glycerol	10.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Nocardia transvalensis*.

Asparagine Broth**Composition per liter:**

DL-Asparagine	30.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g

pH 6.9–7.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix well until dissolved. Adjust pH to between 6.9 and 7.2. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For a presumptive test medium in the differentiation of nonfermentative Gram-negative bacteria, especially *Pseudomonas aeruginosa*.

Aspergillus Differential Medium**Composition per liter:**

Agar	15.0g
Pancreatic digest of casein	15.0g
Yeast extract	10.0g
Ferric citrate	0.5g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 7.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of *Aspergillus flavus*. *Aspergillus flavus* appears as bright orange colonies.

ATB Acid Tomato Broth**Composition per liter:**

Glucose	10.0g
Peptone	10.0g
Yeast Extract	5.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Tomato juice	250.0mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus fructivorans*, *Lactobacillus homohiochii*, *Lactobacillus kefiranoferiens*, and *Leuconostoc oenos*.

Azide Blood Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
NaCl	5.0g
Beef extract	3.0g
NaN ₃	0.2g
Sheep blood, defibrinated	50.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45–50°C. Aseptically add 50.0mL of sterile defibrinated sheep blood. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the isolation and differentiation of streptococci and staphylococci from specimens containing mixed flora.

Azide Blood Agar with Crystal Violet (Packer's Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
NaCl	5.0g
Beef extract	3.0g
NaN ₃	0.9g
Crystal Violet	2.0mg
Sheep blood, defibrinated	50.0mL
pH 7.2 ± 0.2 at 25°C	

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15

psi pressure–121°C. Cool to 45–50°C. Aseptically add 50.0mL of sterile defibrinated sheep blood. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the isolation and enumeration of fecal streptococci from nonclinical specimens such as water and food. Also used for the isolation of *Streptococcus pneumoniae* and *Erysipelothrix rhusiopathiae*.

Azide Broth

(Azide Glucose Broth)

(Azide Dextrose Broth)

Composition per liter:

Pancreatic digest of casein	15.0g
Glucose	7.5g
Beef extract	4.5g
NaCl	7.5g
NaN ₃	0.2g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Prepare double-strength broth for samples larger than 1.0mL.

Use: Used in the multiple-tube technique as a presumptive test for the presence of fecal streptococci.

Azide Medium

Composition per liter:

Peptone	10.0g
K ₂ HPO ₄	5.0g
Glucose	5.0g
NaCl	5.0g
Yeast extract	3.0g
KH ₂ PO ₄	2.0g
NaN ₃	0.25g
Bromcresol Purple solution	2.0mL
pH 7.2 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 10.0mL:

Bromcresol Purple	0.16g
Ethanol	10.0mL

Preparation of Bromcresol Purple Solution: Add Bromcresol Purple to ethanol and bring volume to 10.0mL. Mix thoroughly.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Streptococcus* species and *Staphylococcus* species.

B₁₂ Culture Agar, USP

Composition per liter:

Agar	15.0g
Glucose	10.0g
Proteose peptone No. 3	7.5g
Yeast extract	7.5g
KH ₂ PO ₄	2.0g
Polysorbate 80	0.1g
Tomato juice	100.0mL
pH 6.8 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes in an upright position.

Use: For the cultivation and maintenance of *Lactobacillus leishmannii* ATCC 7830 to be used as the test organism in the Vitamin B₁₂ assay according to the USP.

B₁₂ Inoculum Broth, USP

Composition per liter:

Glucose	10.0g
Proteose peptone No. 3	7.5g
Yeast extract	7.5g
K ₂ HPO ₄	2.0g
Polysorbate 80	0.1g
Tomato juice	100.0mL
pH 6.8 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the preparation of inoculum cultures of *Lactobacillus leishmannii* ATCC 7830, which is used as the test organism in the vitamin B₁₂ assay according to the USP.

Bacillus Agar

Composition per liter:

Agar	20.0g
(NH ₄) ₂ SO ₄	1.3g
Glucose	1.0g
Yeast extract	1.0g
KH ₂ PO ₄	0.37g
MgSO ₄ ·7H ₂ O	0.25g
CaCl ₂ ·2H ₂ O	0.07g
FeCl ₃	0.02g

pH 4.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 3.5. Prepare a separate agar solution by adding 20.0g/500.0mL of distilled/deionized water. Autoclave solutions separately for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically combine both solutions. This procedure avoids acid hydrolysis of the agar. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of acidophilic *Bacillus* species such as *Bacillus acidocaldarius*.

Bacillus Agar, Modified

Composition per liter:

Agar	20.0g
(NH ₄) ₂ SO ₄	0.2g
Glucose	1.0g
Yeast extract	1.0g
KH ₂ PO ₄	0.6g
MgSO ₄ ·7H ₂ O	0.5g
CaCl ₂ ·2H ₂ O	0.25g

pH 3.0–4.0 at 25°C

Preparation of Medium: Add components, except agar and glucose, to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 3.5. Prepare a separate agar and glucose solution by adding 20.0g of agar and 1.0g of glucose to 500.0mL of distilled/deionized water. Autoclave solutions separately for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically combine both solutions. This procedure avoids acid hydrolysis of the agar. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of acidophilic *Bacillus* species such as *Bacillus acidocaldarius*.

Bacillus Agar, 1/4 Strength

Composition per liter:

Agar	18.0g
Yeast extract	2.5g
Pancreatic digest of casein	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus megaterium*.

Bacillus Broth, 1/4 Strength

Composition per liter:

Yeast extract	2.5g
Pancreatic digest of casein	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus megaterium*.

***Bacillus cereus* Medium (BCM)**

Composition per 110.0mL:

Agar	2.0g
D-Mannitol	1.0g
(NH ₄) ₂ PO ₄	0.1g
KCl	0.02g
MgSO ₄ ·7H ₂ O	0.02g
Yeast extract	0.02g
Bromcresol Purple	4.0mg
Egg yolk emulsion, 20%	10.0mL
pH 7.0 ± 0.2 at 25°C	

Egg Yolk Emulsion, 20%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	80.0mL

Preparation of Egg Yolk Emulsion, 20%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 20.0mL of egg yolk emulsion and add to 80.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components—except egg yolk emulsion, 20%—to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile egg yolk emulsion, 20%. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Bacillus cereus*.

***Bacillus cereus* Selective Agar Base**

Composition per liter:

Agar	15.0g
Sodium pyruvate	10.0g
Mannitol	10.0g
Na ₂ HPO ₄	2.5g
NaCl	2.0g
Peptone	1.0g
KH ₂ PO ₄	0.25g
Bromthymol Blue	0.12g
MgSO ₄ ·7H ₂ O	0.1g
Egg yolk emulsion	25.0mL
Polymyxin B solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Egg Yolk Emulsion:

Composition:

Chicken egg yolks	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Polymyxin B Solution:

Composition per 10.0mL:

Polymyxin B	100,000U

Preparation of Polymyxin B Solution: Add Polymyxin B to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except egg yolk emulsion and polymyxin B solution, to distilled/deionized water and bring volume to 965.0mL. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add sterile polymyxin B and 25.0mL of sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the selection and presumptive identification of *Bacillus cereus*. Also for the isolation and enumeration of these bacteria. *Bacillus cereus* grows as moderate-sized (5mm) crenated colonies, which are turquoise, surrounded by a precipitate of egg yolk, which is also turquoise.

***Bacillus coagulans* Medium**

Composition per liter:

Agar	20.0g
Glucose	5.0g

Proteose peptone	5.0g
Yeast extract	5.0g
K ₂ HPO ₄	4.0g
MnSO ₄ ·4H ₂ O solution	10.0mL
CaCl ₂ solution	10.0mL
pH 5.0 ± 0.2 at 25°C	

MnSO₄·4H₂O Solution:**Composition per 10.0mL:**

MnSO ₄ ·4H ₂ O	0.05mg

Preparation of MnSO₄·4H₂O Solution: Add MnSO₄·4H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

CaCl₂ Solution:**Composition per 10.0mL:**

CaCl ₂	0.045mg

Preparation of CaCl₂ Solution: Add CaCl₂ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except MnSO₄·4H₂O solution and CaCl₂ solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Avoid overheating. Cool to 45°–50°C. Aseptically add sterile MnSO₄·4H₂O solution and CaCl₂ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Bacillus coagulans*.

Bacillus Medium**Composition per liter:**

Agar	25.0g
Peptone	6.0g
Pancreatic digest of casein	3.0g
Yeast extract	3.0g
Beef extract	1.5g
MnSO ₄ ·4H ₂ O	1.0μg

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Bacillus* species.

Bacillus fastidiosus Agar**Composition per liter:**

Allantoin	20.0g
Agar	15.0g
K ₂ HPO ₄	0.8g

MgSO ₄ ·7H ₂ O	0.5g
KH ₂ PO ₄	0.2g
CaCl ₂ ·2H ₂ O	50.0mg
FeSO ₄ ·7H ₂ O	10.0mg
MnSO ₄ ·4H ₂ O	1.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Bacillus fastidiosus*.

Bacillus fastidiosus Medium**Composition per liter:**

Agar	15.0g
Na ₂ HPO ₄ ·12H ₂ O	6.0g
Yeast extract	2.5g
Uric acid	1.0g
Mineral solution	100.0mL

pH 7.0 ± 0.2 at 25°C

Mineral Solution:**Composition per 100.0mL:**

KH ₂ PO ₄	0.1g
MgSO ₄ ·7H ₂ O	0.03g
CaCl ₂	0.01g
NaCl	0.01g
FeCl ₃ ·6H ₂ O	1.0mg

Preparation of Mineral Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Bacillus fastidiosus*.

Bacillus Medium**Composition per liter:**

(NH ₄) ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.2g
KCl	0.2g
Yeast extract	0.2g
Glucose solution	50.0mL
Bromcresol Purple solution	15.0mL

pH 7.0 ± 0.2 at 25°C

Glucose Solution:**Composition per 100.0mL:**

Glucose	10.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute 9.5mL volumes into test tubes that contain an inverted Durham tube. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 0.5mL of sterile glucose to each tube. Mix thoroughly.

Use: For cultivation and differentiation of *Bacillus* species based on acid and gas production from glucose.

Bacillus stearothermophilus Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Yeast extract	5.0g
K ₂ HPO ₄	2.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus stearothermophilus*.

Bacillus stearothermophilus Defined Broth

Composition per 100.0mL:

Mineral salts solution	10.0mL
Potassium phosphate buffer	5.0mL
L-Glutamate·HCl (1% solution)	4.0mL
L-Leucine (1% solution)	1.64mL
L-Lysine·HCl (1% solution)	1.4mL
L-Serine (1% solution)	1.4mL
L-Aspartate (1% solution)	1.3mL
L-Valine (1% solution)	1.26mL
Biotin (0.01% solution)	1.0mL
Glucose (20% solution)	1.0mL
L-Isoleucine (1% solution)	1.0mL
L-Proline (1% solution)	1.0mL
Nicotinic acid (0.01% solution)	1.0mL
Thiamine·HCl (0.01% solution)	1.0mL
L-Phenylalanine (1% solution)	0.86mL
L-Alanine (1% solution)	0.84mL
L-Threonine (1% solution)	0.84mL
L-Arginine·HCl (1% solution)	0.64mL
L-Tyrosine (1% solution)	0.56mL
L-Methionine (1% solution)	0.52mL
Glycine (1% solution)	0.5mL
L-Asparagine·H ₂ O (1% solution)	0.5mL

L-Cystine (1% solution)	0.5mL
L-Glutamine (1% solution)	0.5mL
L-Histidine·HCl·H ₂ O (1% solution)	0.42mL
L-Tryptophan (1% solution)	0.3mL
CaCl ₂ (5% solution)	0.01mL
FeCl ₃ ·6H ₂ O (0.05% solution)	0.01mL
MnCl ₂ (10mm solution)	0.01mL
ZnSO ₄ ·7H ₂ O (5% solution)	0.01mL
pH 7.3 ± 0.2 at 25°C	

Mineral Salts Solution:

Composition per liter:

NaCl	10.0g
NH ₄ Cl	10.0g
MgSO ₄	4.0g

Preparation of Mineral Salts Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Potassium Phosphate Buffer:

Composition per 500.0mL:

K ₂ HPO ₄	125.0g
KH ₂ PO ₄	30.0g

Preparation of Potassium Phosphate Buffer: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Use: For the cultivation of *Bacillus stearothermophilus* in a chemically defined medium.

Bacillus stearothermophilus Sporulation Broth

Composition per liter:

Agar	20.0g
Pancreatic digest of gelatin	5.0g
Yeast extract	4.0g
Beef extract	3.0g
MnCl ₂ ·4H ₂ O	10.0μg
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and sporulation of *Bacillus stearothermophilus*.

Bacillus thermoantarcticus Medium

Composition per liter:

Yeast extract	6.0g

NaCl.....	3.0g
Soil extract.....	500.0mL
pH 5.6–5.8 at 25°C	

Soil Extract:**Composition** per liter:

Garden soil, air dried	400.0g

Preparation of Soil Extract: Pass 400.0g of air-dried garden soil through a coarse sieve. Add soil to 960.0mL of tap water. Mix thoroughly. Autoclave for 60 min at 15 psi pressure–121°C. Cool to room temperature. Allow residue to settle. Decant supernatant solution. Filter through Whatman filter paper. Distribute into bottles in 200.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Store at room temperature until clear.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.6–5.8. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus thermoantarcticus*.

Bacillus* Xylose Salts*Composition** per liter:

Yeast extract.....	5.0g
Xylose.....	5.0g
NaCl.....	1.0g
NH ₄ Cl.....	1.0g
KH ₂ PO ₄	0.5g
MgSO ₄ ·7H ₂ O.....	0.5g
CaCl ₂ ·2H ₂ O.....	0.05g
Trace mineral solution	10.0mL
Vitamin solution.....	10.0mL
pH 4.0 ± 0.2 at 25°C	

Trace Mineral Solution:**Composition** per liter:

CoCl ₂ ·6H ₂ O.....	0.2g
FeSO ₄ ·7H ₂ O.....	0.13g
ZnCl ₂ ·2H ₂ O.....	0.1g
MnCl ₂ ·4H ₂ O.....	0.1g
CaCl ₂ ·2H ₂ O.....	20.0mg
Na ₂ SeO ₃	20.0mg
Na ₂ WO ₄ ·2H ₂ O.....	20.0mg
NaMoO ₄ ·2H ₂ O.....	1.0mg
H ₃ BO ₃	0.5mg
CuSO ₄ ·5H ₂ O.....	0.4mg
KI.....	0.1mg

Preparation of Trace Mineral Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Vitamin Solution:**Composition** per liter:

Pyridoxine-HCl.....	10.0mg
Thiamine-HCl.....	5.0mg
Riboflavin.....	5.0mg
Nicotinic acid.....	5.0mg
Calcium pantothenate.....	5.0mg
<i>p</i> -Aminobenzoic acid.....	5.0mg
Thioctic acid.....	5.0mg
Biotin.....	2.0mg
Folic acid.....	2.0mg
Cyanocobalamin.....	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH of medium to 4.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Bacillus* species that can utilize xylose as a carbon source.

BAGG Broth**(Buffered Azide Glucose Glycerol Broth)****Composition** per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
Glucose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	4.0g
KH ₂ PO ₄	1.5g
NaN ₃	0.5g
Bromocresol Purple	0.015g
Glycerol.....	5.0mL

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add 5.0mL of glycerol to 900.0mL of distilled/deionized water. Add remaining components and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 10 psi pressure–116°C.

Use: For the cultivation of fecal streptococci. It is recommended for qualitative presumptive and confirmatory tests for fecal streptococci.

Baird-Parker Agar**Composition per liter:**

Agar	17.0g
Glycine	12.0g
Sodium pyruvate	10.0g
Pancreatic digest of casein	10.0g
Beef extract	5.0g
LiCl	5.0g
Yeast extract	1.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: Used as a base for the preparation of egg-tellurite-glycine-pyruvate agar for the selective isolation and enumeration of coagulase-positive staphylococci from foods.

Baird-Parker Agar**Composition per liter:**

Agar	17.0g
Glycine	12.0g
Sodium pyruvate	10.0g
Pancreatic digest of casein	10.0g
Beef extract	5.0g
LiCl	5.0g
Yeast extract	1.0g
Sulfamethazine solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Sulfamethazine Solution:**Composition per 10.0mL:**

Sulfamethazine	0.05g

Preparation of Sulfamethazine Solution: Add sulfamethazine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sulfamethazine solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile sulfamethazine solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: Used as a base for the preparation of egg-tellurite-glycine-pyruvate agar for the selective isolation and enumeration of coagulase-positive staphylococci from foods.

Baird-Parker Agar, Supplemented**Composition per liter:**

Agar	17.0g
Glycine	12.0g
Sodium pyruvate	10.0g
Pancreatic digest of casein	10.0g
Beef extract	5.0g
LiCl	5.0g
Yeast extract	1.0g
RPF supplement	100.0mL

pH 7.0 ± 0.2 at 25°C

RPF Supplement:**Composition per 100.0mL:**

Bovine fibrinogen	3.75g
Trypsin inhibitor	25.0mg
K ₂ TeO ₃	25.0mg
Rabbit plasma	25.0mL

Caution: Potassium tellurite is toxic.

Preparation of RPF Supplement: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except RPF supplement, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of filter-sterilized RPF supplement. Mix thoroughly but gently. Pour into sterile Petri dishes.

Use: For the selective isolation and enumeration of coagulase-positive staphylococci from foods.

Balamuth Medium**Composition per 200.0mL:**

Dehydrated egg yolk	36.0g
Dried liver concentrate	1.0g
Rice starch	0.2g
Potassium phosphate buffer, pH 7.5	125.0mL
NaCl solution	125.0mL

pH 7.3 ± 0.2 at 25°C

NaCl Solution**Composition per 200.0mL:**

NaCl	1.6g

Preparation of NaCl Solution: Add NaCl to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly.

Potassium Phosphate Buffer, 0.067M**Composition per 200.0mL:**

K ₂ HPO ₄ (1M solution)	8.6mL
KH ₂ PO ₄ (1M solution)	4.66mL

Preparation of Potassium Phosphate Buffer:

Combine the K_2HPO_4 and KH_2PO_4 solutions. Bring volume to 200.0mL with distilled/deionized water. Adjust pH to 7.5.

Preparation of Medium: Add dehydrated egg yolk to 36.0mL of distilled/deionized water. Add 125.0mL of 0.8% NaCl. Mix thoroughly in a blender. Heat in a covered, double boiler until infusion reaches 80°C and maintain at this temperature for 20 min. Add 20.0mL of distilled/deionized H_2O . Filter through a layer of cheesecloth. To 90–100.0mL of filtrate add 0.8% NaCl solution to bring volume to 125.0mL. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 4°C. Filter. To filtrate, add an equal volume of 0.067M potassium phosphate buffer, pH 7.5. Add 1.0g of dried liver concentrate. Mix thoroughly. Distribute into tubes or flasks in 10.0mL volumes. Autoclave for 20 min at 15 psi pressure–121°C. Prior to inoculation, add 0.01g of rice starch to each tube.

Use: For the cultivation and maintenance of *Entamoeba histolytica* and other intestinal protozoa.

Baird-Parker Medium (BAM M17)

Composition per liter:

Agar	20.0g
Glycine	12.0g
Sodium pyruvate	10.0g
Pancreatic digest of casein	10.0g
Beef extract	5.0g
$LiCl \cdot 6H_2O$	5.0g
Yeast extract	1.0g
EY tellurite enrichment	50.0mL
pH 7.0 ± 0.2 at 25°C	

EY Tellurite Enrichment:

Composition per 100.0mL:

Chicken egg yolks	10
K_2TeO_3	0.15g
NaCl (0.9% solution)	50.0mL

Preparation of EY Tellurite Enrichment:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack 11 eggs and separate yolks from whites. Mix egg yolks. Measure 30.0mL of egg yolk emulsion and add to 70.0mL of 0.9% NaCl solution. Mix thoroughly. Add 0.15g K_2TeO_3 . Filter sterilize. Warm to 45°–50°C.

Caution: Potassium tellurite is toxic.

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except EY tellurite enrichment, to distilled/deionized

water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48°–50°C. Aseptically add 50.0mL of sterile EY tellurite enrichment. Mix thoroughly. Pour into sterile Petri dishes. The medium must be densely opaque. Dry plates before use. Plates can be stored for up to 5 days at 20–25°C before use.

Use: For the selective isolation and enumeration of coagulase-positive staphylococci from foods.

Basal Synthetic Medium

Composition per liter:

L-Glutamic acid	20.0g
$(NH_4)_2SO_4$	4.0g
K_2HPO_4	1.88g
KH_2PO_4	0.57g
$MgSO_4 \cdot 7H_2O$	0.2g
Salt solution	10.0mL

Salt Solution:

Composition per liter:

$FeCl_3 \cdot 6H_2O$	0.6g
$MnCl_2 \cdot 4H_2O$	0.6g
$ZnCl_2$	0.6g
$CuSO_4 \cdot 5H_2O$	0.6g
$CaCl_2 \cdot 2H_2O$	0.6g
NaCl	0.6g

Preparation of Salt Solution: Add components to 1.0L of distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Acinetobacter lwoffii*.

Base Layer Agar with Nutrient Overlay Agar

Composition per 2.5L:

Fat substrate	50.0g
Nutrient agar	1.5L
Basal medium	1.0L

Fat Substrate:

Composition:

Fat	50.0g

Preparation of Fat Substrate: Tributyrin, corn oil, soybean oil, any cooking oil, lard, tallow, or triglycerides that do not contain antioxidants or other inhibitory substances may be used. Remove free fatty acids in the fat substrate by dissolving 50.0g of fat substrate in 500.0mL of petroleum ether. Pass the solution through an activated alumina column. Remove

the petroleum ether by evaporation on a steam table under 100% N₂. Autoclave for 30 min at 15 psi pressure–121°C. Cool to 50°C.

Nutrient Agar:

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g

Preparation of Nutrient Agar: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Source: The medium is available as a premixed powder from BD Diagnostic Systems.

Basal Medium:

Composition per liter:

Agar	15.0g
Victoria Blue B solution	200.0mL

Preparation of Basal Medium: Add agar to 800.0mL of distilled/deionized water. If tributyrin is used as the fat substrate, add agar to 1.0L of distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. If tributyrin is not used as the fat substrate, aseptically add 200.0mL of Victoria Blue B solution. Mix thoroughly.

Victoria Blue B Solution:

Composition per 200.0mL:

Victoria Blue B	0.12g

Preparation of Victoria Blue B Solution: Add the Victoria Blue B solution to 200.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Aseptically combine 1.0L of sterile basal medium with 50.0g of sterile fat substrate in a warm, sterile blender container. Blend for 1 min until homogenized. Rapidly pour into sterile Petri dishes in 7.0mL volumes. Dry the surface of the plates by partially opening the lids in a laminar flow hood for 15 min. Add dilution of food samples to be tested. When the inoculum is dry, pour nutrient agar as an overlay onto each plate. Use 10–12mL of nutrient agar per plate.

Use: For the isolation, cultivation, and identification of lipolytic microorganisms from food.

BC Motility Medium (*Bacillus cereus* Motility Medium)

Composition per liter:

Pancreatic digest of casein	10.0g

Glucose	5.0g
Agar	3.0g
Na ₂ HPO ₄	2.5g
Yeast extract	2.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 2.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and observation of motility of *Bacillus cereus*.

BCM *Bacillus cereus* Group Plating Medium

Composition per liter:

Proprietary

Source: This medium is available from Biosynth International, Inc.

Use: For detection of *Bacillus cereus* in foods. The medium contains 5-bromo-4-chloro-3-indoxyl-myoinositol-1-phosphate, which changes from colorless to turquoise upon enzymatic cleavage. *B. cereus*, *B. mycoides*, *B. thuringiensis*, and *B. weihenstephanensis* secrete phosphatidylinositol phospholipase C and so grow as turquoise colonies with species-specific morphologies.

BCM O157:H7(+) Plating Medium

Composition per liter:

Proprietary

Source: This medium is available from Biosynth International, Inc.

Use: For detection of this highly pathogenic EHEC serovar BCM O157:H7(+).

BCP Azide Broth (Bromocresol Purple Azide Broth)

Composition per liter:

Casein peptone	10.0g
Yeast extract	10.0g
D-Glucose	5.0g
NaCl	5.0g
K ₂ HPO ₄	2.7g
KH ₂ PO ₄	2.7g
NaN ₃	0.5g
Bromocresol Purple	0.032g
pH 6.9 ± 0.2 at 25°C	

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For use in the confirmation test for the presence of fecal streptococci.

BCP D Agar

(Bromcresol Purple Deoxycholate Agar)

Composition per liter:

Agar	25.0g
Lactose	10.0g
Sucrose	10.0g
Pancreatic digest of casein	7.5g
Thiopeptone	7.5g
NaCl	5.0g
Yeast extract	2.0g
Sodium citrate	2.0g
Sodium deoxycholate	1.0g
Bromcresol Purple	0.02g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Pour into sterile Petri dishes without sterilization. Do not autoclave. Use the same day.

Use: For the isolation, cultivation, and differentiation of Gram-negative enteric bacilli. For the isolation and cultivation of *Salmonella*, *Shigella*, and other nonlactose- and nonsucrose-fermenting microorganisms. Nonlactose/nonsucrose fermenting microorganisms appear as colorless or blue colonies. Lactose/sucrose-fermenting microorganisms, such as coliform bacteria, appear as yellow-opaque white colonies surrounded by a zone of precipitated deoxycholate.

BCP DCLS Agar

(Bromcresol Purple Deoxycholate Citrate Lactose Sucrose Agar)

Composition per liter:

Agar	14.0g
Sodium citrate	10.0g
Lactose	7.5g
Sucrose	7.5g
Pancreatic digest of casein	7.5g
Peptone	7.5g
NaCl	5.0g
Na ₂ S ₂ O ₃ ·5H ₂ O	5.0g
Yeast extract	3.0g
Meat extract	3.0g
Sodium deoxycholate	2.5g
Bromcresol Purple	0.02g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Pour into sterile Petri dishes without sterilization. Do not autoclave. Use the same day.

Use: For the differential isolation of Gram-negative enteric bacilli. For the isolation of *Salmonella*, *Shigella*, and other nonlactose- and nonsucrose-fermenting microorganisms. Nonlactose/nonsucrose-fermenting microorganisms appear as colorless or blue colonies. Lactose/sucrose-fermenting microorganisms, such as coliform bacteria, appear as yellow-opaque white colonies surrounded by a zone of precipitated deoxycholate.

Beef Extract Broth

Composition per liter:

Peptone	5.0g
Beef extract	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat gently and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a wide variety of microorganisms. Recommended for the culture of microorganisms from milk.

Beef Infusion Agar

Composition per liter:

Ground defatted beef	453.6g
Agar	20.0g
Peptone	10.0g
NaCl	5.0g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Add ground beef to 1.0L of distilled/deionized water. Let stand overnight at 4°C. Gently heat and bring to 80°–90°C for 60 min. Let stand for 2 hr. Filter through muslin. To filtrate, add peptone and salt. Mix thoroughly. Adjust pH to 7.6 with 4% NaOH. Filter through Whatman #1 filter paper. Bring volume of filtrate to 1.0L. Add agar. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of microorganisms.

BG Sulfa Agar

(Brilliant Green Sulfapyridine Agar)

Composition per liter:

Agar	20.0g
Proteose peptone No. 3	10.0g

Lactose	10.0g
Sucrose	10.0g
NaCl	5.0g
Yeast extract	3.0g
Sodium sulfapyridine	1.0g
Brilliant Green	0.125g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat gently to boiling. Distribute into tubes or flasks. Autoclave for no longer than 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes if desired.

Use: For the selective isolation of *Salmonella* species other than *Salmonella typhi* from food, dairy products, eggs and egg products, and feed. *Salmonella* appear as red, pink, or white colonies surrounded by zones of bright red.

Bicarbonate Agar

Composition per 100.0mL:

Soybean-casein digest agar	90.0mL
Sodium bicarbonate solution	10.0mL

pH 7.3 ± 0.2 at 25°C

Soybean-Casein Digest Agar:

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	15.0g
Papaic digest of soybean meal	5.0g
NaCl	5.0g

Preparation of Soybean-Casein Digest Agar:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C.

Sodium Bicarbonate Solution:

Composition per 10.0mL:

NaHCO ₃	0.7g

Preparation of Sodium Bicarbonate Solution:

Add NaHCO₃ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: To 90.0mL of cooled, sterile soybean-casein digest agar, aseptically add 10.0mL of sterile sodium bicarbonate solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of *Vibrio* species from foods.

Bile Esculin Agar

Composition per liter:

Oxgall	20.0g
Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
Esculin	1.0g
Ferric citrate	0.5g
Horse serum	50.0mL

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid and BD Diagnostic Systems.

Preparation of Medium: Add components, except horse serum, to distilled/deionized water and bring volume to 950.0L. Mix thoroughly and heat with frequent agitation until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of filter sterilized horse serum. Distribute into sterile Petri dishes or test tubes. Cool tubes in a slanted position.

Use: For differentiation between group D streptococci and nongroup D streptococci. To differentiate members of the Enterobacteriaceae, particularly *Klebsiella*, *Enterobacter*, and *Serratia*, from other enteric bacteria. To differentiate *Listeria monocytogenes*. Bile tolerance and esculin hydrolysis (seen as a dark brown to black complex) are presumptive for enterococci (group D streptococci).

Bile Esculin Agar (BAM M18)

Composition per liter:

Oxgall	40.0g
Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
Esculin	1.0g
Ferric citrate	0.5g

pH 6.6 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Distribute into sterile Petri dishes or test tubes. Cool tubes in a slanted position.

Use: For differentiation between group D streptococci and nongroup D streptococci. To differentiate members of the Enterobacteriaceae, particularly *Klebsiella*, *Enterobacter*, and *Serratia*, from other enteric bacteria. To differentiate *Listeria monocytogenes*. Bile tolerance and esculin hydrolysis (seen as a dark brown to black complex) are presumptive for enterococci (group D streptococci).

Bile Oxalate Sorbose Broth (BOS Broth)

Composition per liter:

Na ₂ HPO ₄	9.14g
Sodium oxalate	5.0g
Bile salts.....	2.0g
NaCl.....	1.0g
CaCl ₂ ·2H ₂ O.....	0.01g
MgSO ₄ ·7H ₂ O	0.01g
Asparagine solution	100.0mL
Methionine solution	100.0mL
Sorbose solution.....	100.0mL
Yeast extract solution	10.0mL
Sodium pyruvate solution	10.0mL
Metanil Yellow solution.....	10.0mL
Sodium nitrofurantoin solution.....	10.0mL
Irgasan® solution.....	1.0mL

pH 7.6 ± 0.2 at 25°C

Asparagine Solution:

Composition per 100.0mL:

Asparagine	1.0g

Preparation of Asparagine Solution: Add asparagine to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Methionine Solution:

Composition per 100.0mL:

Methionine	1.0g

Preparation of Methionine Solution: Add methionine to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Sorbose Solution:

Composition per 100.0mL:

Sorbose.....	10.0g

Preparation of Sorbose Solution: Add sorbose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Yeast Extract Solution:

Composition per 10.0mL:

Yeast extract.....	0.025g

Preparation of Yeast Extract Solution: Add yeast extract to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Sodium Pyruvate Solution:

Composition per 10.0mL:

Sodium pyruvate	0.05g

Preparation of Sodium Pyruvate Solution: Add sodium pyruvate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Metanil Yellow Solution:

Composition per 10.0mL:

Metanil Yellow	0.025g

Preparation of Metanil Yellow Solution: Add Metanil Yellow to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Sodium Nitrofurantoin Solution:

Composition per 10.0mL:

Sodium nitrofurantoin.....	0.01g

Preparation of Sodium Nitrofurantoin Solution: Add sodium nitrofurantoin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Irgasan Solution:

Composition per 10.0mL:

Irgasan.....	0.04g
Ethanol (95% solution)	10.0mL

Preparation of Irgasan Solution: Add Irgasan to 10.0mL of ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except asparagine solution, methionine solution, sorbose solution, yeast extract solution, sodium pyruvate solution, Metanil Yellow solution, sodium nitrofurantoin solution, and Irgasan solution, to distilled/deionized water and bring volume to 659.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile asparagine solution, 100.0mL of sterile methionine solution, 100.0mL of sterile sorbose solution, 10.0mL of sterile yeast extract solution, 10.0mL of sterile sodium pyruvate solution, 10.0mL of sterile Metanil Yellow solution, 10.0mL of sterile sodium nitrofurantoin solution, and 1.0mL of sterile Irgasan solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Yersinia enterocolitica* from foods.

Bile Salts Brilliant Green Starch Agar (BBGS Agar)

Composition per liter:

Agar	15.0g
Soluble starch.....	10.0g
Proteose peptone.....	10.0g
Beef extract.....	5.0g
Bile salts.....	5.0g
Brilliant Green (0.05% solution)	1.0mL

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave

for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Aeromonas hydrophila* from foods.

Bile Salts Gelatin Agar

Composition per 100.0mL:

Gelatin.....	3.0g
Agar	1.5g
Pancreatic digest of casein.....	1.0g
NaCl.....	1.0g
Sodium taurocholate	0.5g
Na ₂ CO ₃	0.1g
Water	100.0mL

pH 8.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Vibrio cholerae*.

Biosynth Chromogenic Medium for *Listeria monocytogenes* (BCM for *Listeria monocytogenes*) (BAM M17a)

Composition per liter:

Proprietary

Source: This medium is available from Biosynth International, Inc.

Use: To differentiate *Listeria monocytogenes* and *L. ivanovii* from other *Listeria* spp. The supplements render the medium selective. Differential activity for all *Listeria* species is based upon a chromogenic substrate included in the medium. This is a complete test system with a fluorogenic selective enrichment broth and a chromogenic plating medium both detecting the virulence factor Phosphatidylinositol specific Phospholipase C (PI-PLC). The medium contains a substrate for phosphatidylinositol-specific phospholipase C (PlcA) enzymes. The selective enrichment broth is fluorogenic. The plating medium for rapid detection and enumeration of pathogenic *Listeria* combines cleavage of the chromogenic PI-PLC substrate with the additional production of a white precipitate surrounding the target colonies.

Biotin Assay Medium

Composition per liter:

Glucose	40.0g
Sodium acetate	20.0g

Vitamin assay casamino acids	12.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
DL-Tryptophan.....	0.2g
L-Cystine	0.2g
Adenine sulfate	0.02g
FeSO ₄	0.02g
Guanine·HCl	0.02g
MgSO ₄ ·7H ₂ O	0.02g
NaCl.....	0.02g
Uracil	0.02g
Calcium pantothenate	2.0mg
Niacin.....	2.0mg
Pyridoxine·HCl	2.0mg
Riboflavin	2.0mg
Thiamine·HCl	2.0mg
p-Aminobenzoic acid.....	0.2mg

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 5.0mL volumes. Add standard solution or test solutions to each tube. Adjust the volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For use in the microbiological assay of biotin using *Lactobacillus plantarum* as the test microorganism.

Bismuth Sulfite Agar

Composition per liter:

Agar	20.0g
Bi ₂ (SO ₃) ₃	8.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
Beef extract.....	5.0g
Glucose	5.0g
Na ₂ HPO ₄	4.0g
FeSO ₄ ·7H ₂ O.....	0.3g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until boiling. Boil for 1 min. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes while gently shaking flask to disperse precipitate. Use plates the same day as prepared.

Use: For the selective isolation and identification of *Salmonella typhi* and other enteric bacilli. *Salmonella typhi* appears as flat, black, “rabbit-eye” colonies surrounded by a zone of black with a metallic sheen.

Bismuth Sulfite Agar Wilson and Blair (BAM M19)

Composition per liter:

Agar	20.0g
Pancreatic digest of casein	10.0g
Bi ₂ (SO ₃) ₃	8.0g
Beef extract	5.0g
Glucose	5.0g
Na ₂ HPO ₄	4.0g
FeSO ₄ ·7H ₂ O	0.3g
Brilliant Green	0.025g
pH 7.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until boiling. Boil for 1 min. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes while gently shaking flask to disperse precipitate. Let plates dry for about 2 hr with lids partially removed. Use plates the within one day of preparation; medium loses selectivity after 48 hr.

Use: For the selective isolation and identification of *Salmonella typhi* and other enteric bacilli. *Salmonella typhi* appears as flat, black, “rabbit-eye” colonies surrounded by a zone of black with a metallic sheen.

Bismuth Sulfite Broth (m-Bismuth Sulfite Broth)

Composition per liter:

Bi ₂ (SO ₃) ₃	16.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
Beef extract	10.0g
Glucose	10.0g
Na ₂ HPO ₄	8.0g
FeSO ₄ ·7H ₂ O	0.6g
pH 7.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until boiling. Boil for 1 min. Do not autoclave. Cool to 45°–50°C. Mix to disperse the precipitate and aseptically distribute into sterile tubes or flasks. Use 2.0–2.2mL of medium for each membrane filter.

Use: For the selective isolation of *Salmonella typhi* and other enteric bacilli and for the detection of *Salmonella* by the membrane filter method.

BL Agar (Glucose Blood Liver Agar)

Composition per liter:

Agar	15.0g
Glucose	10.0g
Proteose peptone No. 3	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
Meat extract	3.0g
Phytone™	3.0g
Tween 80	1.0g
Soluble starch	0.5g
Liver extract	150.0mL
Horse blood	50.0mL
L-Cysteine-HCl solution	10.0mL
Solution A	10.0mL
Solution B	5.0mL
pH 7.2 ± 0.2 at 25°C	

Liver Extract:

Composition per 170.0mL:

Liver powder	10.0g

Preparation of Liver Extract: Add 10.0g of liver powder to 170mL of distilled/deionized water. Gently heat to 60°C. Maintain at 50°–60°C for 1 hr. Gently bring to boiling. Boil for 5 min. Adjust pH to 7.2. Filter through Whatman #2 filter paper.

L-Cysteine-HCl Solution:

Composition per 10.0mL:

L-Cysteine-HCl	0.5g

Preparation of L-Cysteine-HCl Solution: Dissolve 0.5g of L-cysteine-HCl in distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Solution A:

Composition per 100.0mL:

K ₂ HPO ₄	10.0g
KH ₂ PO ₄	10.0g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution B:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O	4.0g
NaCl	0.2g
FeSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·H ₂ O	0.2g

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Preparation of Medium: Add components, except liver extract, horse blood, L-cysteine·HCl solution, solution A, and solution B, to distilled/deionized water and bring volume to 775.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 150.0mL of sterile liver extract, 50.0mL of sterile horse blood, 10.0mL of sterile L-cysteine·HCl solution, 10.0mL of sterile solution A, and 5.0mL of sterile solution B. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Clostridium* species, numerous *Lactobacillus* species, *Lactococcus lactis*, *Leuconostoc lactis*, *Leuconostoc mesenteroides*, and *Propionibacterium thoenii*.

Blood Agar (BAM M20)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	15.0g
Papaic digest of soybean meal	5.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 50.0mL of sterile sheep blood. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of fastidious microorganisms.

Blood Agar Base (BAM M20)

Composition per liter:

Agar	15.0g
Beef extract	10.0g
Peptone	10.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–

121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of streptococci and other fastidious microorganisms.

Blood Agar Base (BAM M20a)

Composition per liter:

Beef heart, infusion from	500.0g
Agar	15.0g
Tryptose	10.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of staphylococci, streptococci, and other fastidious microorganisms.

Blood Agar Base (Infusion Agar) (BAM M20)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	13.0g
NaCl	5.0g
Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of streptococci and other fastidious microorganisms.

Blood Agar Base (Infusion Agar) (BAM M21)

Composition per liter:

Heart muscle, infusion from	375.0g
Agar	15.0g
Pancreatic digest of casein	10.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of streptococci and other fastidious microorganisms.

Blood Agar with 1% NaCl (BAM M20)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	15.0g
NaCl	10.0g
Papaic digest of soybean meal	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 50.0mL of sterile sheep blood. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Vibrio hollisae*.

Blood Agar Base No. 2 (BAM M22)

Composition per 1004mL:

Agar	12.0g
Protease peptone	15.0g
NaCl	5.0g
Yeast extract	5.0g
Liver digest	2.5g
Horse blood, defibrinated	50.0mL
FBP solution	4.0mL
pH 7.4 ± 0.2 at 25°C	

FBP Solution:

Composition per 30.0mL:

FeSO ₄	0.25g
NaHSO ₃	0.25g
Sodium pyruvate	0.25g

Preparation of FBP Solution: Add components to distilled/deionized water and bring volume to 30.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse blood and FBP solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48°C. Aseptically add 50.0mL of sterile horse blood. Mix thoroughly. Aseptically add 4.0mL sterile FBP solution. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Brucella* spp. and other fastidious bacteria.

Bolton Broth

Composition per 530mL:

Bolton selective enrichment broth base	500.0mL
Horse blood, lysed	25.0mL
Bolton selective supplement solution	5.0mL
pH 7.4 ± 0.2 at 25°C	

Bolton Selective Enrichment Broth Base:

Composition per liter:

Peptone	10.0g
Lactalbumin hydrolysate	5.0g
Yeast extract	5.0g
NaCl	5.0g
a-ketoglutarate	1.0g
Na-pyruvate	0.5g
Na-metabisulfite	0.5g
Na ₂ CO ₃	0.6g
Hemin	0.01g

Preparation of Bolton Selective Enrichment Broth Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Bolton Broth Supplement Solution:

Composition per 5.0mL

Vancomycin	10.0mg
Cefoperazone	10.0mg
Trimethoprim	10.0mg
Cycloheximide	10.0mg
Ethanol	2.5mL

Preparation of Bolton Supplement Solution: Add antibiotics to 2.5mL ethanol. Mix thoroughly.

Bring volume to 5.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine 500.0mL warm Bolton selective enrichment broth base, 25.0mL lysed horse blood, and 5.0mL Bolton selective supplement solution.

Use: For the enrichment of *Campylobacter* spp. from foods.

Bouillon Medium

Composition per liter:

Peptone.....	15.0g
Meat extract	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	5.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat with frequent agitation and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the general cultivation of heterotrophic microorganisms.

Bovine Albumin Tween 80 Medium, Ellinghausen and McCullough, Modified (Albumin Fatty Acid Broth, *Leptospira* Medium)

Composition per liter:

Basal medium	900.0mL
Albumin fatty acid supplement.....	100.0mL

Basal Medium:

Composition per liter:

Na ₂ HPO ₄ , anhydrous	1.0g
NaCl.....	1.0g
KH ₂ PO ₄ , anhydrous.....	0.3g
NH ₄ Cl (25% solution).....	1.0mL
Glycerol (10% solution).....	1.0mL
Sodium pyruvate (10% solution).....	1.0mL
Thiamine·HCl (0.5% solution).....	1.0mL
pH 7.4 ± 0.2 at 25°C	

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Albumin Fatty Acid Supplement:

Composition per 200.0mL:

Bovine albumin fraction V	20.0g
Polysorbate (Tween) 80 (10% solution)	25.0mL

FeSO ₄ ·7H ₂ O (0.5% solution).....	20.0mL
CaCl ₂ ·2H ₂ O (1.5% solution).....	2.0mL
MgCl ₂ ·2H ₂ O (1.5% solution)	2.0mL
Vitamin B ₁₂ (0.2% solution)	2.0mL
ZnSO ₄ ·7H ₂ O (0.4% solution)	2.0mL
CuSO ₄ ·5H ₂ O (0.3% solution)	0.2mL

Preparation of Albumin Fatty Acid Supplement: Add bovine albumin to 100.0mL of distilled/deionized water. Mix thoroughly. Add remaining components while stirring. Adjust pH to 7.4. Bring volume to 200.0mL with distilled/deionized water. Filter sterilize. Store at –20°C.

Preparation of Medium: Aseptically combine 100.0mL of sterile albumin fatty acid supplement and 900.0mL of sterile basal medium. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leptospira* species.

Bovine Albumin Tween 80 Semisolid Medium, Ellinghausen and McCullough, Modified (Albumin Fatty Acid Semisolid Medium, Modified)

Composition per liter:

Basal medium	900.0mL
Albumin fatty acid supplement.....	100.0mL

Basal Medium:

Composition per liter:

Agar	2.2g
Na ₂ HPO ₄ , anhydrous	1.0g
NaCl.....	1.0g
KH ₂ PO ₄ , anhydrous.....	0.3g
NH ₄ Cl (25% solution)	1.0mL
Glycerol (10% solution)	1.0mL
Sodium pyruvate (10% solution).....	1.0mL
Thiamine·HCl (0.5% solution)	1.0mL
pH 7.4 ± 0.2 at 25°C	

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Albumin Fatty Acid Supplement:

Composition per 200.0mL:

Bovine albumin fraction V	20.0g
Polysorbate (Tween) 80 (10% solution)	25.0mL
FeSO ₄ ·7H ₂ O (0.5% solution).....	20.0mL
CaCl ₂ ·2H ₂ O (1.5% solution).....	2.0mL
MgCl ₂ ·2H ₂ O (1.5% solution)	2.0mL
Vitamin B ₁₂ (0.2% solution)	2.0mL
ZnSO ₄ ·7H ₂ O (0.4% solution)	2.0mL
CuSO ₄ ·5H ₂ O (0.3% solution)	0.2mL

Preparation of Albumin Fatty Acid Supplement: Add bovine albumin to 100.0mL of distilled/deionized water. Mix thoroughly. Add remaining components while stirring. Adjust pH to 7.4. Bring volume to 200.0mL with distilled/deionized water. Filter sterilize. Store at -20°C .

Preparation of Medium: Aseptically combine 100.0mL of sterile albumin fatty acid supplement and 900.0mL of sterile basal medium. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leptospira* species.

Bovine Serum Albumin Tween 80 Agar (BSA Tween 80 Agar)

Composition per liter:

Basal medium	900.0mL
Albumin supplement	100.0mL

Basal Medium:

Composition per liter:

Agar	11.0g
Na_2HPO_4	1.0g
NaCl	1.0g
KH_2PO_4	0.3g
Glycerol (10% solution)	1.0mL
NH_4Cl (25% solution)	1.0mL
Sodium pyruvate (10% solution)	1.0mL
Thiamine (0.5% solution)	1.0mL

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 25°C .

Albumin Supplement:

Composition per 100.0mL:

Bovine albumin	10.0g
Tween 80 (10% solution)	12.5mL
FeSO_4 (0.5% solution)	10.0mL
$\text{MgCl}_2\text{-CaCl}_2$ solution	1.0mL
Cyanocobalamin (0.02% solution)	1.0mL
ZnSO_4 (0.4% solution)	1.0mL

Preparation of Albumin Supplement: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Adjust pH to 7.4. Filter sterilize.

$\text{MgCl}_2\text{-CaCl}_2$ Solution:

Composition per 100.0mL:

$\text{CaCl}_2\cdot 2\text{H}_2\text{O}$	1.5g
$\text{MgCl}_2\cdot 6\text{H}_2\text{O}$	1.5g

Preparation of $\text{MgCl}_2\text{-CaCl}_2$ Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: To 900.0mL of cooled, sterile basal medium, aseptically add 100.0mL of sterile albumin supplement. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Leptospira* species.

Bovine Serum Albumin Tween 80 Broth (BSA Tween 80 Broth)

Composition per liter:

Basal medium	900.0mL
Albumin supplement	100.0mL
pH 7.4 ± 0.2 at 25°C	

Basal Medium:

Composition per liter:

Na_2HPO_4	1.0g
NaCl	1.0g
KH_2PO_4	0.3g
Glycerol (10% solution)	1.0mL
NH_4Cl (25% solution)	1.0mL
Sodium pyruvate (10% solution)	1.0mL
Thiamine (0.5% solution)	1.0mL

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 25°C .

Albumin Supplement:

Composition per 100.0mL:

Bovine albumin	10.0g
Tween 80 (10% solution)	12.5mL
FeSO_4 (0.5% solution)	10.0mL
$\text{MgCl}_2\text{-CaCl}_2$ solution	1.0mL
Cyanocobalamin (0.02% solution)	1.0mL
ZnSO_4 (0.4% solution)	1.0mL

Preparation of Albumin Supplement: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Adjust pH to 7.4. Filter sterilize.

$\text{MgCl}_2\text{-CaCl}_2$ Solution:

Composition per 100.0mL:

$\text{CaCl}_2\cdot 2\text{H}_2\text{O}$	1.5g
$\text{MgCl}_2\cdot 6\text{H}_2\text{O}$	1.5g

Preparation of $\text{MgCl}_2\text{-CaCl}_2$ Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: To 900.0mL of cooled, sterile basal medium, aseptically add 100.0mL of sterile albumin supplement. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and cultivation of *Leptospira* species.

**Bovine Serum Albumin
Tween 80 Soft Agar
(BSA Tween 80 Soft Agar)
(Semisolid BSA Tween 80 Medium)**

Composition per liter:

Basal medium	900.0mL
Albumin supplement.....	100.0mL

Basal Medium:

Composition per liter:

Agar	2.0g
Na ₂ HPO ₄	1.0g
NaCl.....	1.0g
KH ₂ PO ₄	0.3g
Glycerol (10% solution).....	1.0mL
NH ₄ Cl (25% solution).....	1.0mL
Sodium pyruvate (10% solution)	1.0mL
Thiamine (0.5% solution).....	1.0mL

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Albumin Supplement:

Composition per 100.0mL:

Bovine albumin.....	10.0g
Tween 80 (10% solution)	12.5mL
FeSO ₄ (0.5% solution).....	10.0mL
CaCl ₂ -MgCl ₂ solution.....	1.0mL
Cyanocobalamin (0.02% solution)	1.0mL
ZnSO ₄ (0.4% solution).....	1.0mL

Preparation of Albumin Supplement: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Adjust pH to 7.4. Filter sterilize.

MgCl₂-CaCl₂ Solution:

Composition per 100.0mL:

CaCl ₂ ·2H ₂ O.....	1.5g
MgCl ₂ ·6H ₂ O	1.5g

Preparation of MgCl₂-CaCl₂ Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: To 900.0mL of cooled, sterile basal medium, aseptically add 100.0mL of sterile albumin supplement. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leptospira* species.

Brain Heart Infusion Agar

Composition per liter:

Pancreatic digest of casein.....	16.0g
Agar	13.5g
Brain heart, solids from infusion	8.0g
Peptic digest of animal tissue	5.0g
NaCl.....	5.0g
Na ₂ HPO ₄	2.5g
Glucose	2.0g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks while shaking to distribute precipitate. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of fastidious microorganisms, including bacteria, yeasts, and molds.

**Brain Heart Infusion Agar
(BHI Agar)
(BAM M24 Medium 1)**

Composition per liter:

Beef heart infusion.....	250.0g
Calf brain infusion	200.0g
Agar	13.5g
Proteose peptone.....	10.0g
NaCl.....	5.0g
Na ₂ HPO ₄ ·12H ₂ O.....	2.5g
Glucose	2.0g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of fastidious and nonfastidious aerobic and anaerobic microorganisms.

**Brain Heart Infusion Agar
(BAM M24 Medium 2)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	14.5g
Brain heart, solids from infusion	6.0g
Peptic digest of animal tissue	6.0g
NaCl.....	5.0g

Glucose	3.0g
Na ₂ HPO ₄	2.5g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks while shaking to distribute precipitate. Autoclave for 15 min at 15 psi pressure–121°C. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of a wide variety of fastidious microorganisms, including bacteria, yeasts, and molds.

Brain Heart Infusion Agar 0.7%
(BHI Agar 0.7%)
(BAM M23)

Composition per liter:

Beef heart infusion	250.0g
Calf brain infusion	200.0g
Proteose peptone	10.0g
Agar	7.0g
NaCl	5.0g
Na ₂ HPO ₄ ·12H ₂ O	2.5g
Glucose	2.0g
pH 5.3 ± 0.2 at 25°C	

Source: This medium without agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except agar, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.3 with 1N HCl. Mix thoroughly. Add agar. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the detection of staphylococcal enterotoxin.

Brain Heart Infusion Agar 0.7%
(BHI Agar 0.7%)
(BAM M23)

Composition per liter:

Pancreatic digest of gelatin	14.5g
Agar	7.0g
Brain heart, solids from infusion	6.0g
Peptic digest of animal tissue	6.0g
NaCl	5.0g
Glucose	3.0g
Na ₂ HPO ₄	2.5g
pH 5.3 ± 0.2 at 25°C	

Source: This medium without agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except agar, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.3 with 1N HCl. Mix thoroughly. Add agar. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the detection of staphylococcal enterotoxin.

Brain Heart Infusion Broth
(BHI Broth)
(BAM M24 Medium 1)

Composition per liter:

Beef heart infusion	250.0g
Calf brain infusion	200.0g
Proteose peptone	10.0g
NaCl	5.0g
Na ₂ HPO ₄ ·12H ₂ O	2.5g
Glucose	2.0g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of fastidious and nonfastidious aerobic and anaerobic microorganisms.

Brain Heart Infusion Broth
(BHI Broth)
(BAM M24 Medium 2)

Composition per liter:

Pancreatic digest of gelatin	14.5g
Brain heart, solids from infusion	6.0g
Peptic digest of animal tissue	6.0g
NaCl	5.0g
Glucose	3.0g
Na ₂ HPO ₄	2.5g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks while shaking to distribute precipitate. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of microorganisms, including bacteria, yeasts, and molds, especially fastidious species.

Brewer Anaerobic Agar**Composition per liter:**

Agar	20.0g
Proteose peptone No. 3	10.0g
Glucose	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
NaCl	5.0g
Sodium thioglycolate	2.0g
Sodium formaldehyde sulfoxylate	1.0g
Resazurin	2.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of anaerobic and microaerophilic microorganisms.

Brewer Thioglycolate Medium**Composition per liter:**

Beef, infusion from	500.0g
Proteose peptone	10.0g
NaCl	5.0g
Glucose	5.0g
K ₂ HPO ₄	2.0g
Sodium thioglycolate	0.5g
Agar	0.5g
Methylene Blue	2.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of anaerobic and microaerophilic microorganisms. For testing the sterility of biological products and materials.

Brigg's Liver Tomato Broth**Composition per liter:**

Glucose	20.0g
Neopeptone	15.0g
Yeast extract	6.0g
NaCl	5.0g
Tween 80	1.0g
Soluble starch	0.5g
L-Cysteine·HCl	0.2g

Tomato juice	400.0mL
Liver extract	75.0mL

pH 5.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. (Note: 3.0g of proteolysed liver may be used instead of the liver extract.) Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus acetotolerans*, a bacterium found in fermented vinegar broth.

Brilliant Green Agar**Composition per liter:**

Agar	20.0g
Lactose	10.0g
Sucrose	10.0g
Peptic digest of animal tissue	5.0g
Pancreatic digest of casein	5.0g
NaCl	5.0g
Phenol Red	0.08g
Brilliant Green	0.0125g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the selective isolation of *Salmonella* other than *Salmonella typhi* from food and dairy products. *Salmonella* other than *Salmonella typhi* appear as red/pink/white colonies surrounded by a zone of red in the agar, indicating nonlactose/sucrose fermentation. *Proteus* or *Pseudomonas* species may appear as small red colonies. Lactose- or sucrose-fermenting bacteria appear as yellow-green colonies surrounded by a zone of yellow-green in the agar.

Brilliant Green Agar, Modified**Composition per liter:**

Agar	12.0g
Lactose	10.0g
Sucrose	10.0g
Beef extract	5.0g
Peptone	5.0g
NaCl	5.0g
Yeast extract	3.0g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g

Phenol Red	0.09g
Brilliant Green	4.7mg
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Addition of 1.0g of sodium sulfacetamide and 250.0mg of sodium mandelate enhances inhibition of contaminating microorganisms. Pour into sterile Petri dishes.

Use: For the selective isolation of *Salmonella* other than *Salmonella typhi* from food and dairy products. *Salmonella* other than *Salmonella typhi* appear as red/pink/white colonies surrounded by a zone of red in the agar, indicating nonlactose/sucrose fermentation. *Proteus* or *Pseudomonas* species may appear as small red colonies. Lactose- or sucrose-fermenting bacteria appear as yellow-green colonies surrounded by a zone of yellow-green in the agar.

Brilliant Green Agar, Modified (Edel-Kampelmacher Medium)

Composition per liter:

Agar	12.0g
Peptone.....	10.0g
Lactose	10.0g
Sucrose.....	10.0g
Lab-Lemco.....	5.0g
Yeast extract.....	3.0g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g
Phenol Red.....	0.09g
Brilliant Green	0.0047g
Sulphamandate supplement	10.0mL
pH 6.9 ± 0.2 at 25°C	

Sulphamandate Supplement:

Composition per 10.0mL:

Sodium sulphacetamide.....	1.0g
Sodium mandelate.....	0.25g

Preparation of Sulphamandate Supplement: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to a boil to dissolve the components completely. Do not autoclave. Cool to 50°C. Aseptically add 10.0mL

sulphamandate supplement. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation and differentiation of *Salmonella* spp. other than *S. typhi*.

Brilliant Green Agar with Sulfadiazine

Composition per liter:

Agar	20.0g
Lactose	10.0g
Sucrose.....	10.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
NaCl.....	5.0g
Yeast extract.....	3.0g
Phenol Red.....	0.08g
Sulfadiazine	0.08g
Brilliant Green	0.0125g
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the selective detection of *Salmonella* in foods, especially from egg products. *Salmonella* other than *Salmonella typhi* appear as red/pink colonies surrounded by a zone of red in the agar indicating nonlactose/sucrose fermentation. *Proteus* or *Pseudomonas* species may appear as small red colonies. Lactose- or sucrose-fermenting bacteria appear as yellow-green colonies surrounded by a zone of yellow-green in the agar.

Brilliant Green Bile Agar

Composition per liter:

Noble agar.....	10.15g
Pancreatic digest of gelatin.....	8.25g
Lactose.....	1.9g
Na ₂ SO ₃	0.205g
Basic Fuchsin.....	0.078g
Erioglaucine.....	0.065g
FeCl ₃	0.0295g
KH ₂ PO ₄	0.015g
Oxgall, dehydrated.....	2.95mg
Brilliant Green	0.03mg
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Basic Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contamination of the skin.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. For plating 10.0mL samples, prepare the medium double strength. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes. Care should be taken to avoid exposure of the prepared medium to light.

Use: For the detection and enumeration of coliform bacteria in materials of sanitary importance such as water, sewage, and foods. *Escherichia coli* appears as dark red colonies with a pink halo. *Enterobacter* species appear as pink colonies.

**Brilliant Green 2%-Bile Broth,
Fluorocult®
(Fluorocult Brilliant Green
2%-Bile Broth)
(BRILA)**

Composition per liter:

Ox bile, dried	20.0g
Peptone.....	10.0g
Lactose.....	10.0g
L-Tryptophan.....	1.0g
4-Methylumbelliferyl-β-D-glucuronide	0.1g
Brilliant Green	0.0133g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool. Distribute into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C. Do not autoclave longer. The prepared broth is clear and green.

Use: For the cultivation of *Escherichia coli*. Bile and brilliant green almost completely inhibit the growth of undesired microbial flora, in particular Gram-positive microorganisms. *E. coli* shows a positive fluorescence under UV light (366 nm). A positive indole reaction and, if necessary gas formation due to fermenting lactose, confirm the findings.

**Brilliant Green Bile Broth
(Brilliant Green Lactose Bile Broth)**

Composition per liter:

Oxgall, dehydrated.....	20.0g
Lactose	10.0g

Pancreatic digest of gelatin.....	10.0g
Brilliant Green	0.0133g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing inverted Durham tubes, in 10.0mL amounts for testing 1.0mL or less of sample. Autoclave for 12 min (not longer than 15 min) at 15 psi pressure–121°C. After sterilization, cool the broth rapidly. Medium is sensitive to light.

Use: For the detection of coliform microorganisms in foods, dairy products, water, and wastewater as well as in other materials of sanitary importance. Turbidity in the broth and gas in the Durham tube are positive indications of *Escherichia coli*.

**Brilliant Green Broth
(m-Brilliant Green Broth)**

Composition per liter:

Proteose peptone No. 3	20.0g
Lactose	20.0g
Sucrose.....	20.0g
NaCl.....	10.0g
Yeast extract.....	6.0g
Phenol Red.....	0.16g
Brilliant Green	0.025g
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with frequent mixing. Boil for 1 min. Cool to 25°C. Add 2.0mL to each sterile absorbent filter used.

Use: For the selective isolation and differentiation of *Salmonella* spp. by the membrane filter method.

**Brilliant Green Lactose Bile Broth
(BAM M25)**

Composition per liter:

Oxgall, dehydrated.....	20.0g
Lactose	10.0g
Pancreatic digest of gelatin.....	10.0g
Brilliant Green	0.0133g
pH 7.2 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add lactose and pancreatic digest of gelatin to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Add 20.0g oxgall dissolved in 200.0mL distilled/deionized water. The pH of this solution should be 7.0–7.5. Mix thoroughly. Bring volume to 975.0mL with distilled/deionized water. Adjust pH to 7.4. Add 13.3mL of 0.1% aqueous Brilliant Green in distilled/deionized water. Adjust volume to 1.0L with distilled/deionized water. Distribute into tubes containing inverted Durham tubes, in 10.0mL amounts for testing 1.0mL or less of sample. Make sure that the fluid level covers the inverted vials. Autoclave for 15 min at 15 psi pressure–121°C. After sterilization, cool the broth rapidly. Medium is sensitive to light. The final pH should be 7.2 ± 0.1 at 25°C.

Use: For the detection of coliform microorganisms in foods, dairy products, water, and wastewater as well as in other materials of sanitary importance. Turbidity in the broth and gas in the Durham tube are positive indications of *Escherichia coli*.

Brilliant Green Bile Broth with MUG

Composition per liter:

Oxgall, dehydrated.....	20.0g
Lactose	10.0g
Pancreatic digest of gelatin.....	10.0g
MUG (4-Methyl umbelliferyl- β-D-glucuronide).....	0.05g
Brilliant Green	0.013g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing inverted Durham tubes, in 10.0mL amounts for testing 1.0mL or less of sample. Autoclave for 12 min (not longer than 15 min) at 15 psi pressure–121°C. After sterilization, cool the broth rapidly.

Use: For the detection of coliform microorganisms in foods, dairy products, water, and wastewater, as well as in other materials of sanitary importance. The presence of *Escherichia coli* and other coliforms is determined by the presence of fluorescence in the tube.

Brilliant Green Phenol Red Agar

Composition per liter:

Agar	15.0g
Lactose	15.0g
Peptone.....	10.0g
Meat extract	5.0g
NaCl.....	5.0g

Phenol Red.....	0.08g
Brilliant Green.....	0.0125g
pH 6.9 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Salmonella* species.

Bromcresol Purple Broth

Composition per liter:

Peptone	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Bromcresol Purple	0.04g
Carbohydrate solution.....	10.0mL
pH 7.0 ± 0.2 at 25°C	

Carbohydrate Solution:

Composition per 10.0mL:

Carbohydrate.....	5.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the differentiation of a variety of microorganisms based on their fermentation of specific carbohydrates. Bacteria that ferment the specific carbohydrate turn the medium yellow. When bacteria produce gas, the gas is trapped in the Durham tube.

Bromcresol Purple Broth (BAM M26)

Composition per liter:

Peptone	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Bromcresol Purple	0.04g
Carbohydrate solution.....	50.0mL
pH 7.0 ± 0.2 at 25°C	

Carbohydrate Solution:

Composition per 50.0mL:

Carbohydrate.....	25.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 10 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 50.0mL of carbohydrate solution. Aseptically distribute into tubes or flasks. Alternately distribute the medium without the carbohydrate solution into test tubes that contain an inverted Durham tube prior to autoclaving. Then autoclave for 10 min at 15 psi pressure–121°C, cool to 25°C, and aseptically add the carbohydrate solution to each tube to yield a final carbohydrate concentration of 5%.

Use: For the differentiation of a variety of microorganisms based on their fermentation of specific carbohydrates. Bacteria that ferment the specific carbohydrate turn the medium yellow. When bacteria produce gas, the gas is trapped in the Durham tube.

Bromcresol Purple Broth with NaCl (BAM M26)

Composition per liter:

NaCl.....	25.0g
Peptone.....	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Bromcresol Purple	0.04g
Carbohydrate solution.....	50.0mL
pH 7.0 ± 0.2 at 25°C	

Carbohydrate Solution:

Composition per 50.0mL:

Carbohydrate.....	25.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 10 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 50.0mL of carbohydrate solution. Aseptically distribute into tubes or flasks. Alternately distribute the medium without the carbohydrate solution into test tubes that contain an inverted Durham tube prior to autoclaving. Then autoclave for 10 min at 15 psi pressure–121°C, cool to 25°C, and aseptically add the carbohydrate solution to each tube to yield a final carbohydrate concentration of 5%.

Use: For the differentiation of a halophilic *Vibrio* spp. Bacteria that ferment the specific carbohydrate turn the medium yellow. When bacteria produce gas, the gas is trapped in the Durham tube.

Bromcresol Purple Dextrose Broth (BCP Broth) (BAM M27)

Composition per liter:

Glucose	10.0g
Peptone	5.0g
Beef extract.....	3.0g
Bromcresol Purple solution	2.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 10.0mL:

Bromcresol Purple	0.16g
Ethanol (95% solution)	10.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to 10.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 12–15mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to ferment glucose. Bacteria that ferment glucose turn the medium yellow.

Bromthymol Blue Agar

Composition per liter:

Agar	11.0g
Peptone	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Lactose (33% solution)	27.0mL
Bromthymol Blue (1% solution)	10.0mL
Sodium thiosulfate (50% solution)	2.0mL
Glucose (33% solution)	1.2mL
Maranil solution (5% solution)	1.0mL
pH 7.7–7.8 at 25°C	

Preparation of Medium: Add agar, peptone, NaCl, and yeast extract to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.0. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 45°–50°C. Filter sterilize the lactose solution, Bromthymol Blue solution, sodium thiosulfate solution, glucose solution, and maranil solution separately. To the cooled, sterile agar solution, aseptically add 27.0mL of sterile lactose solution, 10.0mL of sterile Bromthymol Blue solution, 2.0mL of sterile sodium

thiosulfate solution, 1.2mL of sterile glucose solution, and 1.0mL of sterile maranil solution. Mix thoroughly. Adjust pH to 7.7–7.8. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of members of the Enterobacteriaceae.

***Brucella* Agar**

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose	1.0g
NaHSO ₃	0.1g
Horse blood, defibrinated	100.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Heat gently with frequent mixing. Boil for 1 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Add 100.0mL of sterile defibrinated horse blood. Mix gently and pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Brucella* species. For the isolation and cultivation of nonfastidious and fastidious microorganisms from a variety of specimens.

***Brucella* Agar with Vitamin K₁**

Composition per liter:

Agar	17.5g
Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose	1.0g
NaHSO ₃	0.1g
Sheep blood, defibrinated	50.0mL
Vitamin K ₁ solution	1.0mL
pH 7.0 ± 0.2 at 25°C	

Vitamin K₁ Solution:

Composition per 20.0mL:

Vitamin K ₁	0.2g
Ethanol, absolute.....	20.0mL

Preparation of Vitamin K₁ Solution: Add vitamin K₁ to 20.0mL of ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sheep blood and vitamin K₁ solution, to distilled/deionized water and bring volume to 949.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile sheep blood and 1.0mL of sterile vitamin K₁ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Brucella* species.

***Brucella* Albimi Broth with 0.16% Agar**

Composition per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Agar	1.6g
Glucose	1.0g
NaHSO ₃	0.1g
Horse blood, defibrinated	100.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except horse blood, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Heat gently with frequent mixing. Boil for 1 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile defibrinated horse blood. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter* species.

***Brucella* Albimi Broth with 0.16% Agar and 1% Glycine (ATCC Medium 2161)**

Composition per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
Glycine.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Agar	1.6g
Glucose	1.0g
NaHSO ₃	0.1g
Horse blood, defibrinated	100.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except horse blood, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Heat gently with frequent mixing. Boil for 1 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile defibrinat-

ed horse blood. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter* species.

Brucella Anaerobic Blood Agar

Composition per liter:

Vitamin K ₁	0.01g
Anaerobic agar base.....	950.0mL
Sheep blood, sterile, defibrinated	50.0mL

Anaerobic Agar Base

Composition per liter:

Pancreatic digest of casein.....	17.5g
Agar	15.0g
Glucose	10.0g
Papaic digest of soybean meal.....	2.5g
NaCl.....	2.5g
Sodium thioglycolate	2.0g
Sodium formaldehyde sulfoxylate.....	1.0g
L-Cystine-HCl-H ₂ O	0.4g
Methylene Blue.....	0.002g

pH 7.0 ± 0.2 at 25°C

Preparation of Anaerobic Agar Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: To 950.0mL of cooled, sterile anaerobic agar base, aseptically add 10.0mg of vitamin K₁ and 50.0mL of sterile, defibrinated sheep blood.

Use: For the isolation of anaerobes.

Brucella Broth

(*Brucella Albimi* Broth)

Composition per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose	1.0g
NaHSO ₃	0.1g
Horse blood, defibrinated	50.0mL

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except horse blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat gently with frequent mixing. Boil for 1 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile horse blood.

Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter coli*, *Campylobacter fecalis*, and *Brucella* species. Also used for the isolation and cultivation of a wide variety of fastidious and nonfastidious microorganisms.

Brucella Broth with 0.16% Agar **(ATCC Medium 1116)**

Composition per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Agar	1.6g
Glucose	1.0g
NaHSO ₃	0.1g
Horse blood, defibrinated	50.0mL

pH 7.0 ± 0.2 at 25°C

Source: This medium without agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except horse blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat gently with frequent mixing. Boil for 1 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile horse blood. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter fetus* subsp. *Fetus* and *Campylobacter jejuni* subsp. *Jejuni*

Brucella Medium Base

Composition per liter:

Agar	15.0g
Glucose	10.0g
Peptone	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g

pH 7.5 ± 0.2 at 25°C

Preparation: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat gently and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation of *Campylobacter* species.

Brucella Medium, Selective

Composition per liter:

Agar	15.0g

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose.....	1.0g
NaHSO ₃	0.1g
Horse serum.....	100.0mL
VCNF antibiotic solution.....	10.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of VCNF Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse serum and VCNF antibiotic solution, to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile horse serum and 10.0mL of VCNF antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and maintenance of *Brucella* species.

***Brucella* Selective Medium**

Composition per liter:

Beef heart, infusion from.....	500.0g
Agar.....	15.0g
Tryptose.....	10.0g
NaCl.....	5.0g
Glucose.....	2.5g
Gelatin.....	1.0g
Sheep blood.....	100.0mL
Antibiotic solution.....	10.0mL

pH 7.4 ± 0.2 at 25°C

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide.....	1.0g
Bacitracin.....	250,000U
Circulin.....	250,000U
Polymyxin B.....	100,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sheep blood and antibiotic solution, to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile sheep blood and 10.0mL of sterile antibiotic solution. Mix

thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Brucella* species.

***Brucella* Semisolid Medium with Cysteine**

Composition per liter:

Peptamin.....	10.0g
Pancreatic digest of casein.....	10.0g
Glycine.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Agar.....	1.8g
Glucose.....	1.0g
L-Cysteine-HCl·H ₂ O.....	0.2g
NaHSO ₃	0.1g
Sodium citrate.....	0.1g
Neutral Red solution.....	10.0mL

pH 7.0 ± 0.2 at 25°C

Neutral Red Solution:

Composition per 100.0mL:

Neutral Red.....	0.2g
Ethanol.....	10.0mL

Preparation of Neutral Red Solution: Add Neutral Red to 10.0mL of ethanol. Bring volume to 100.0mL.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Campylobacter* species based on H₂S production from cysteine.

***Brucella* Semisolid Medium with Glycine**

Composition per liter:

Peptamin.....	10.0g
Pancreatic digest of casein.....	10.0g
Glycine.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Agar.....	1.8g
Glucose.....	1.0g
NaHSO ₃	0.1g
Sodium citrate.....	0.1g
Neutral Red solution.....	10.0mL

pH 7.0 ± 0.2 at 25°C

Neutral Red Solution:**Composition** per 100.0mL:

Neutral Red	0.2g
Ethanol	10.0mL

Preparation of Neutral Red Solution: Add Neutral Red to 10.0mL of ethanol. Bring volume to 100.0mL.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Campylobacter* species based on glycine utilization.

Brucella Semisolid Medium with NaCl**Composition** per liter:

NaCl	35.0g
Peptamin	10.0g
Pancreatic digest of casein	10.0g
Yeast extract	2.0g
Agar	1.8g
Glucose	1.0g
NaHSO ₃	0.1g
Sodium citrate	0.1g
Neutral Red solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Neutral Red Solution:**Composition** per 100.0mL:

Neutral Red	0.2g
Ethanol	10.0mL

Preparation of Neutral Red Solution: Add Neutral Red to 10.0mL of ethanol. Bring volume to 100.0mL.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Campylobacter* species based on glycine utilization.

Brucella Semisolid Medium with Nitrate**Composition** per liter:

Peptamin	10.0g
Pancreatic digest of casein	10.0g
Glycine	10.0g
KNO ₃	10.0g
NaCl	5.0g
Yeast extract	2.0g
Agar	1.8g
Glucose	1.0g

NaHSO ₃	0.1g
Sodium citrate	0.1g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Campylobacter* species based on nitrate reduction.

**Buffered Enrichment Broth
(BAM M52)****Composition** per liter:

Na ₂ HPO ₄	9.6g
KH ₂ PO ₄	1.35g
Pyruvic acid solution	11.1mL
Nalidixic acid solution	8.0mL
Cycloheximide solution	5.0mL
Acriflavin solution	2.0mL
pH 7.3 ± 0.1 at 25°C	

Nalidixic Acid Solution:**Composition** per 10.0mL:

Nalidixic acid, sodium salt	0.05g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin Solution:**Composition** per 10.0mL:

Acriflavin-HCl	0.05g

Preparation of Acriflavin Solution: Add acriflavin-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:**Composition** per 10.0mL:

Cycloheximide	0.1g
Ethanol, 40%	10.0mL

Preparation of Cycloheximide Solution: Add cycloheximide to 40% ethanol and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Pyruvate Solution:**Composition** per 20.0mL:

Na-pyruvate	2.0g

Preparation of Pyruvate Solution: Add Na-pyruvate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except pyruvate solution, nalidixic acid solution, acriflavin solution, and cycloheximide solution, to distilled/deionized water and bring volume to

973.9.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 11.1mL sterile pyruvate solution. Mix thoroughly. Aseptically add 8.0mL sterile nalidixic acid solution, 5.0mL sterile cycloheximide solution, and 2.0mL sterile acriflavin solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of of *Listeria* spp.

Buffered Peptone Water

Composition per liter:

Pancreatic digest of gelatin.....	10.0g
NaCl.....	5.0g
Na ₂ HPO ₄	3.5g
KH ₂ PO ₄	1.5g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: Used as a preenrichment medium for the isolation of *Salmonella*, especially injured microorganisms, from various food sources.

CAL Broth

(Cellobiose Arginine Lysine Broth)

Composition per liter:

L-Arginine·HCl.....	6.5g
L-Lysine·HCl.....	6.5g
NaCl.....	5.0g
Cellobiose.....	3.5g
Yeast extract.....	3.0g
Sodium deoxycholate.....	1.5g
Neutral Red.....	0.03g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat to boiling. Do not autoclave. Distribute into sterile tubes in 6.0–8.0mL volumes.

Use: For the isolation and characterization of *Yersinia enterocolitica* and enumeration of *Yersinia enterocolitica* from liquid foods.

Campylobacter Agar, Blaser's (Blaser's Campylobacter Agar)

Composition per liter:

Campylobacter agar base.....	990.0mL
Supplement B.....	10.0mL
pH 7.4 ± 0.2 at 25°C	

Campylobacter Agar Base:

Composition per liter:

Proteose peptone.....	15.0g
Agar.....	12.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Liver digest.....	2.5g

Source: *Campylobacter* agar base and *Campylobacter* antimicrobial supplement B are available as a premixed powder from BD Diagnostic Systems.

Preparation of Campylobacter Agar Base:

Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Supplement B:

Composition per 10.0mL:

Cephalothin.....	15.0mg
Vancomycin.....	10.0mg
Trimethoprim.....	5.0mg
Amphotericin B.....	2.0mg
Polymyxin B.....	2500U

Preparation of Supplement B: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: Prepare 990.0mL of *Campylobacter* agar base. Autoclave and cool to 45°–50°C. Aseptically add 10.0mL of sterile supplement B. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter jejuni* from foods.

Campylobacter Agar, Skirrow's (Skirrow's Campylobacter Agar)

Composition per liter:

Campylobacter agar base.....	990.0mL
Supplement S.....	10.0mL
pH 7.4 ± 0.2 at 25°C	

Campylobacter Agar Base:

Composition per liter:

Proteose peptone.....	15.0g
Agar.....	12.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Liver digest.....	2.5g

Source: *Campylobacter* agar base and *Campylobacter* antimicrobial supplement S are available as a premixed powder from BD Diagnostic Systems.

Preparation of Campylobacter Agar Base:

Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently

heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Supplement S:

Composition per 10.0mL:

Vancomycin	10.0mg
Trimethoprim	5.0mg
Polymyxin B	2500U

Preparation of Supplement S: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: Prepare 990.0mL of *Campylobacter* agar base. Autoclave and cool to 45°–50°C. Aseptically add 10.0mL of sterile supplement S. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter jejuni* from foods.

Campylobacter Blood-Free Agar Base Modified (CCDA, Modified)

Composition per liter:

Peptone	20.0g
Agar	12.0g
NaCl	5.0g
Activated charcoal	4.0g
Casein hydrolysate	3.0g
Na-deoxycholate	1.0g
Na-pyruvate	0.25g
FeSO ₄	0.25g
Cefoperazone-amphotericin B solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Cefoperazone-Amphotericin B Solution:

Composition per 10.0mL:

Cefoperazone	0.016g
Amphotericin B	0.005g

Preparation of Cefoperazone-Amphotericin B Solution: Add cefoperazone and amphotericin B to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components except cefoperazone-amphotericin B solution to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of cefoperazone-amphotericin B solution. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into tubes or flasks.

Use: For the isolation of *Campylobacter* spp. from foods. The use of *Campylobacter* Blood-Free Selective Agar is specified by the UK Ministry of Agriculture, Fisheries and Food (MAFF) in a validated method for isolation of *Campylobacter* from foods. Am-

photericin largely reduces the growth of yeasts and molds. Cefoperazone especially inhibits Enterobacteriaceae.

Campylobacter Blood-Free Selective Agar

Composition per liter:

Agar	12.0g
Beef extract	10.0g
Peptone	10.0g
Charcoal	4.0g
Casein hydrolysate	3.0g
Sodium deoxycholate	1.0g
Fe ₂ SO ₄ ·H ₂ O	0.25g
Sodium pyruvate	0.25g
Cefoperazone solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Cefoperazone Solution:

Composition per 10.0mL:

Sodium cefoperazone	0.032g

Preparation of Cefoperazone Solution: Add sodium cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cefoperazone solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Add 10.0mL of sterile cefoperazone solution. Addition of 10.0mg/L of amphotericin B improves the selectivity of the medium. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species, especially *Campylobacter jejuni*, *Campylobacter coli*, and *Campylobacter lariidis*.

Campylobacter Charcoal Differential Agar (CCDA)

(Preston Blood-Free Medium)

Composition per liter:

Agar	12.0g
Beef extract	10.0g
Peptone	10.0g
NaCl	5.0g
Charcoal	4.0g
Casein hydrolysate	3.0g
Sodium deoxycholate	1.0g
FeSO ₄	0.25g

Sodium pyruvate	0.25g
Cefoperazone solution	10.0mL
pH 7.5 ± 0.2 at 25°C	

Cefoperazone Solution:**Composition per 10.0mL:**

Sodium cefoperazone	0.032g

Preparation of Cefoperazone Solution: Add sodium cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cefoperazone solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 45° – 50°C . Aseptically add 10.0mL of sterile cefoperazone solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Campylobacter* species.

Campylobacter Enrichment Broth**Composition per liter:**

Beef extract	10.0g
Peptone	10.0g
Yeast extract	6.0g
NaCl	5.0g
Horse blood, laked	50.0mL
FBP solution	4.0mL
Antibiotic solution	4.0mL
pH 7.5 ± 0.2 at 25°C	

Horse Blood, Laked:**Composition per 50.0mL:**

Horse blood, fresh	50.0mL

Preparation of Horse Blood, Laked: Add blood to a sterile polypropylene bottle. Freeze overnight at -20°C . Thaw at 8°C . Refreeze at -20°C . Thaw again at 8°C .

FBP Solution:**Composition per 100.0mL:**

FeSO_4	6.25g
NaHSO_3	6.25g
Sodium pyruvate	6.25g

Preparation of FBP Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Antibiotic Solution:**Composition per 10.0mL:**

Cycloheximide	0.1g
Sodium cefoperazone	0.03g
Trimethoprim lactate	0.0125g
Rifampicin	0.01g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except laked horse blood, FBP solution, and antibiotic solution—to distilled/deionized water and bring volume to 942.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 45° – 50°C . Aseptically add 50.0mL of sterile laked horse blood, 4.0mL of FBP solution, and 4.0mL of antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Campylobacter* species from dairy products.

**Campylobacter Enrichment Broth
(BAM M28a)****Composition per 1016.0mL:**

Peptone	10.0g
Yeast extract	5.0g
Lactalbumin hydrolysate	5.0g
NaCl	5.0g
α -Ketoglutamic acid	1.0g
Sodium pyruvate	0.5g
Na_2CO_3	0.6g
$\text{Na}_2\text{S}_2\text{O}_5$	0.5g
Haemin	0.01g
Cefoperazone solution	4.0mL
Trimethoprim lactate solution	4.0mL
Vancomycin solution	4.0mL
Cycloheximide or amphotericin B solution	4.0mL
pH 7.4 ± 0.2 at 25°C	

Cefoperazone Solution:**Composition per 10.0mL:**

Cefoperazone	0.05g

Preparation of Cefoperazone Solution: Add cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 5 days at 4°C , 14 days at -20°C , and 5 months at -70°C .

Trimethoprim Solution:**Composition per 10.0mL:**

Trimethoprim lactate	0.066g

Preparation of Trimethoprim Solution: Add trimethoprim lactate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Alternately add 0.05g trimethoprim hydrochloride to 3.0mL 0.05N HCl. Heat to 50°C . Stir until dissolved. Add distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 1 year at 4°C .

Vancomycin Solution:**Composition** per 10.0mL:

Vancomycin 0.05g

Preparation of Vancomycin Solution: Add vancomycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 2 months at 4°C.

Cycloheximide Solution:**Composition** per 10.0mL:

Cycloheximide 0.025g

Ethanol 2.0mL

Preparation of Cycloheximide Solution: Add cycloheximide to 2.0mL ethanol to dissolve. Mix thoroughly. Add distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 1 year at 4°C.

Amphotericin B Solution:**Composition** per 10.0mL:

Amphotericin B 0.005g

Preparation of Amphotericin B Solution: Add Amphotericin B to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 1 year at -20°C.

Preparation of Medium: Add components, except antimicrobial solutions, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure-121°C. Cool to 25°C. Aseptically add 4.0mL of sterile cefoperazone solution, 4.0mL of sterile trimethoprim lactate solution, 4.0mL of sterile vancomycin solution, and either 4.0mL of sterile cycloheximide solution or 4.0mL of sterile amphotericin B solution. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes or bottles. Close caps tightly to reduce O₂ absorption. Use within 2 weeks.

Use: For the selective isolation and cultivation of *Campylobacter* species. For the preenrichment of *Campylobacter* spp. in food samples. This medium aids resuscitation of sublethally injured cells and overcomes the damaging effects of food processing.

***Campylobacter* Enrichment Broth (FDA Medium M29)**

Composition per 1024.0mL:

Basal medium 950.0mL

Horse blood, lysed 50.0mL

Cefoperazone solution 8.0mL

FBP solution 4.0mL

Trimethoprim lactate solution 4.0mL

Vancomycin solution 4.0mL

Cycloheximide solution 4.0mL

pH 7.5 ± 0.2 at 25°C

Basal Medium:**Composition** per 950.0mL:

Beef extract 10.0g

Peptone 10.0g

Yeast extract 6.0g

NaCl 5.0g

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure-121°C. Cool to 45°-50°C.

FBP Solution:**Composition** per 100.0mL:FeSO₄·7H₂O 6.25gNa₂S₂O₅ 6.25g

Sodium pyruvate 6.25g

Preparation of FBP: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cefoperazone Solution:**Composition** per 10.0mL:

Cefoperazone 0.037g

Preparation of Cefoperazone Solution: Add cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Trimethoprim Lactate Solution:**Composition** per 10.0mL:

Trimethoprim lactate 0.031g

Preparation of Trimethoprim Lactate Solution: Add trimethoprim lactate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Vancomycin Solution:**Composition** per 10.0mL:

Vancomycin 0.025g

Preparation of Vancomycin Solution: Add vancomycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:**Composition** per 10.0mL:

Cycloheximide 0.025g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 950.0mL of cooled sterile basal medium, aseptically add 50.0mL of lysed (fresh, frozen, and thawed) horse blood, 4.0mL of sterile FBP solution, 8.0mL of sterile cefoperazone solution, 4.0mL of sterile trimethoprim lactate solution, 4.0mL of sterile vancomycin solution, and 4.0mL of sterile cycloheximide solution. Mix thor-

oughly. Aseptically distribute into sterile screw-capped tubes or bottles. Close caps tightly to reduce O₂ absorption. Use within 2 weeks.

Use: For the selective isolation and cultivation of *Campylobacter* species.

***Campylobacter* Enrichment Broth (FDA Medium M29)**

Composition per 1020.0mL:

Basal medium	950.0mL
Horse blood, lysed	50.0mL
FBP solution	4.0mL
Cefoperazone solution	4.0mL
Trimethoprim lactate solution	4.0mL
Vancomycin solution	4.0mL
Cycloheximide solution	4.0mL
pH 7.5 ± 0.2 at 25°C	

Basal Medium:

Composition per 950.0mL:

Beef extract	10.0g
Peptone	10.0g
Yeast extract	6.0g
NaCl	5.0g

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

FBP Solution:

Composition per 100.0mL:

FeSO ₄ ·7H ₂ O	6.25g
Na ₂ S ₂ O ₅	6.25g
Sodium pyruvate	6.25g

Preparation of FBP: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cefoperazone Solution:

Composition per 10.0mL:

Cefoperazone	0.037g

Preparation of Cefoperazone Solution: Add cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Trimethoprim Lactate Solution:

Composition per 10.0mL:

Trimethoprim lactate	0.031g

Preparation of Trimethoprim Lactate Solution: Add trimethoprim lactate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Vancomycin Solution:

Composition per 10.0mL:

Vancomycin	0.025g

Preparation of Vancomycin Solution: Add vancomycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.025g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 950.0mL of cooled, sterile basal medium, aseptically add 50.0mL of lysed (fresh, frozen, and thawed) horse blood, 4.0mL of sterile FBP solution, 4.0mL of sterile cefoperazone solution, 4.0mL of sterile trimethoprim lactate solution, 4.0mL of sterile vancomycin solution, and 4.0mL of sterile cycloheximide solution. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes or bottles. Close caps tightly to reduce O₂ absorption. Use within 2 weeks.

Use: For the selective isolation and cultivation of *Campylobacter* species.

***Campylobacter* Enrichment Broth (FDA Medium M29)**

Composition per 1020.0mL:

Basal medium	950.0mL
Horse blood, lysed	50.0mL
FBP solution	4.0mL
Cefoperazone solution	4.0mL
Trimethoprim lactate solution	4.0mL
Rifampicin solution	4.0mL
Cycloheximide solution	4.0mL
pH 7.5 ± 0.2 at 25°C	

Basal Medium:

Composition per 950.0mL:

Beef extract	10.0g
Peptone	10.0g
Yeast extract	6.0g
NaCl	5.0g

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

FBP Solution:

Composition per 100.0mL:

FeSO ₄ ·7H ₂ O	6.25g
Na ₂ S ₂ O ₅	6.25g
Sodium pyruvate	6.25g

Preparation of FBP: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cefoperazone Solution:**Composition** per 10.0mL:

Cefoperazone 0.037g

Preparation of Cefoperazone Solution: Add cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Trimethoprim Lactate Solution:**Composition** per 10.0mL:

Trimethoprim lactate 0.031g

Preparation of Trimethoprim Lactate Solution: Add trimethoprim lactate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Rifampicin Solution:**Composition** per 100.0mL:

Rifampicin 0.25g

Ethanol, absolute 50.0mL

Preparation of Rifampicin Solution: Add rifampicin to 50.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Cycloheximide Solution:**Composition** per 10.0mL:

Cycloheximide 0.025g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 950.0mL of cooled sterile basal medium, aseptically add 50.0mL of lysed (fresh, frozen, and thawed) horse blood, 4.0mL of sterile FBP solution, 4.0mL of sterile cefoperazone solution, 4.0mL of sterile trimethoprim lactate solution, 4.0mL of sterile rifampicin solution, and 4.0mL of sterile cycloheximide solution. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes or bottles. Close caps tightly to reduce O₂ absorption. Use within 2 weeks.

Use: For the selective isolation and cultivation of *Campylobacter* species from dairy products.

Campylobacter* Enrichment Broth*Composition** per liter:

Beef extract 10.0g

Peptone 10.0g

Yeast extract 6.0g

NaCl 5.0g

Horse blood, laked 50.0mL

FBP solution 4.0mL

Antibiotic solution 4.0mL

pH 7.5 ± 0.2 at 25°C

Horse Blood, Laked:**Composition** per 50.0mL:

Horse blood, fresh 50.0mL

Preparation of Horse Blood, Laked: Add blood to a sterile polypropylene bottle. Freeze overnight at -20°C. Thaw at 8°C. Refreeze at -20°C. Thaw again at 8°C.

FBP Solution:**Composition** per 100.0mL:FeSO₄ 6.25gNaHSO₃ 6.25g

Sodium pyruvate 6.25g

Preparation of FBP Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Antibiotic Solution:**Composition** per 10.0mL:

Cycloheximide 0.1g

Sodium cefoperazone 0.03g

Trimethoprim lactate 0.0125g

Vancomycin 0.01g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except horse blood, FBP solution, and antibiotic solution—to distilled/deionized water and bring volume to 942.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile horse blood, 4.0mL of FBP solution, and 4.0mL of antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Campylobacter* species from foods.

Campylobacter fecalis* Medium*Composition** per 1133.6mL:

Starch, soluble 2.4g

Yeast extract 2.4g

Blood agar base 1.0L

Bovine blood 120.0mL

Antibiotic solution 10.0mL

Sodium lactate (60% syrup) 3.6mL

pH 6.8 ± 0.2 at 25°C

Blood Agar Base:**Composition** per liter:

Beef heart, solids from infusion 500.0g

Agar 15.0g

Tryptose 10.0g

NaCl 5.0g

Preparation of Blood Agar Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling.

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide.....	0.12g
Albamycin.....	6.0mg
Bacitracin.....	6000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 1.0L of blood agar base, add soluble starch, yeast extract, and sodium lactate. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile bovine blood and antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and isolation of *Campylobacter fecalis*.

Campylobacter fetus Medium

Composition per liter:

Proteose peptone.....	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Bovine blood.....	50.0mL
Antibiotic solution.....	10.0mL
pH 7.2–7.4 at 25°C	

Antibiotic Solution:

Composition per 10.0mL:

Novobiocin.....	2.0mg
Bacitracin.....	2000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except bovine blood and antibiotic solution, to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile bovine blood and antibiotic solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and cultivation of *Campylobacter fetus*.

Campylobacter fetus Medium

Composition per 1160.0mL:

Fluid thioglycolate agar.....	1.0L

Sheep blood, defibrinated.....	150.0mL
Antibiotic solution.....	10.0mL
pH 7.1 ± 0.2 at 25°C	

Fluid Thioglycolate Agar:

Composition per liter:

Agar.....	15.0g
Pancreatic digest of casein.....	15.0g
Glucose.....	5.5g
Yeast extract.....	5.0g
NaCl.....	2.5g
Agar.....	0.75g
L-Cystine.....	0.5g
Sodium thioglycolate.....	0.5g
Resazurin.....	1.0mg

Preparation of Fluid Thioglycolate Agar:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide.....	0.05g
Novobiocin.....	5.0mg
Bacitracin.....	25,000U
Polymyxin B sulfate.....	10,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 1.0L of cooled, sterile, fluid thioglycolate agar, aseptically add 150.0mL of sterile sheep blood and 10.0mL of sterile antibiotic solution.

Use: For the isolation and cultivation of *Campylobacter fetus* from human specimens.

Campylobacter fetus Selective Medium

Composition per liter:

Fluid thioglycolate agar.....	1.0L
Sheep blood, defibrinated.....	150.0mL
Antibiotic solution.....	10.0mL

Fluid Thioglycolate Agar:

Composition per liter:

Agar.....	15.0g
Pancreatic digest of casein.....	15.0g
Glucose.....	5.5g
Yeast extract.....	5.0g
NaCl.....	2.5g
Agar.....	0.75g
L-Cystine.....	0.5g
Sodium thioglycolate.....	0.5g
Resazurin.....	1.0mg

Preparation of Fluid Thioglycolate Agar:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Antibiotic Solution:**Composition per 10.0mL:**

Cycloheximide.....	0.05g
Cephalothin.....	0.02g
Novobiocin.....	5.0mg
Bacitracin.....	25,000U
Colistin.....	10,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 1.0L of cooled, sterile, fluid thioglycolate agar, aseptically add 150.0mL of sterile sheep blood and 10.0mL of sterile antibiotic solution.

Use: For the isolation and cultivation of *Campylobacter fetus*.

Campylobacter Isolation Agar A**Composition per liter:**

Agar.....	12.0g
Beef extract.....	10.0g
Peptone.....	10.0g
NaCl.....	5.0g
Charcoal.....	4.0g
Casein hydrolysate.....	3.0g
Yeast extract.....	2.0g
Sodium deoxycholate.....	1.0g
FeSO ₄	0.25g
Sodium pyruvate.....	0.25g
Antibiotic solution.....	10.0mL

pH 7.4 ± 0.2 at 25°C

Antibiotic Solution:**Composition per 10.0mL:**

Cycloheximide.....	0.1g
Sodium cefoperazone.....	0.03g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes. Swirl flask while pouring to distribute charcoal.

Use: For the isolation and cultivation of *Campylobacter* species.

***Campylobacter Isolation Agar B*
(Campy Cefex Agar)****Composition per liter:**

Agar.....	15.0g
Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose.....	1.0g
FeSO ₄	0.5g
Sodium pyruvate.....	0.5g
NaHSO ₃	0.35g
Horse blood, laked.....	50.0mL
Antibiotic solution.....	10.0mL

pH 7.0 ± 0.2 at 25°C

Horse Blood, Laked:**Composition per 50.0mL:**

Horse blood, fresh.....	50.0mL

Preparation of Horse Blood, Laked: Add blood to a sterile polypropylene bottle. Freeze overnight at –20°C. Thaw at 8°C. Refreeze at –20°C. Thaw again at 8°C.

Antibiotic Solution:**Composition per 10.0mL:**

Cycloheximide.....	0.1g
Sodium cefoperazone.....	0.033g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse blood and antibiotic solution, to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile horse blood and antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Campylobacter* species.

Campylobacter Medium**Composition per liter:**

Sodium aspartate.....	10.0g
MgSO ₄ ·7H ₂ O.....	1.0g
K ₂ HPO ₄	0.75g
Yeast extract.....	0.2g
CaCl ₂ ·2H ₂ O.....	28.0mg
Resazurin.....	1.0mg

Phosphate-cysteine solution.....	100.0mL
Trace elements solution SL-10	1.0mL
pH 7.0 ± 0.2 at 25°C	

Phosphate-Cysteine Solution:**Composition per 100.0mL:**

NaH ₂ PO ₄	0.25g
L-Cysteine·HCl	0.25g

Preparation of Phosphate-Cysteine Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Trace Elements Solution SL-10:**Composition per liter:**

FeCl ₂ ·4H ₂ O	1.5g
CoCl ₂ ·6H ₂ O	190.0mg
MnCl ₂ ·4H ₂ O	100.0mg
ZnCl ₂	70.0mg
Na ₂ MoO ₄ ·2H ₂ O	36.0mg
NiCl ₂ ·6H ₂ O	24.0mg
H ₃ BO ₃	6.0mg
CuCl ₂ ·2H ₂ O	2.0mg
HCl (25% solution).....	10.0mL

Preparation of Trace Elements Solution SL-10:

Add FeCl₂·4H₂O to 10.0mL of HCl solution. Mix thoroughly. Add distilled/deionized water and bring volume to 1.0L. Add remaining components. Mix thoroughly.

Preparation of Medium: Add components, except phosphate-cysteine solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 100.0mL of sterile phosphate-cysteine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter* species.

**Campylobacter Selective
Medium, Blaser-Wang
(Blaser-Wang Campylobacter Medium)
(Blaser's Agar)
(Campy BAP Medium)**

Composition per liter:

<i>Brucella</i> agar base.....	890.0mL
Sheep blood.....	100.0mL
Antibiotic supplement.....	10.0mL

Brucella Agar Base:**Composition per 890.0mL:**

Agar	15.0g
Glucose	10.0g
Pancreatic digest of casein.....	10.0g

NaCl.....	5.0g
Peptic digest of animal tissue	5.0g
pH 7.5 ± 0.2 at 25°C	

Preparation of Brucella Agar Base: Add components to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:**Composition per 10.0mL:**

Cephalothin.....	15.0mg
Vancomycin	10.0mg
Trimethoprim	5.0mg
Amphotericin B	2.0mg
Polymyxin B.....	2500U

Preparation of Antibiotic Supplement: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: Prepare 890.0mL of *Brucella* agar base. Sterilize as directed. Cool to 50°–55°C and add 100.0mL of sheep blood or 50.0–70.0mL of laked horse blood. Laked blood is prepared by freezing whole blood overnight and thawing to room temperature. Aseptically add 10.0mL of sterile antibiotic supplement. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species.

**Campylobacter Selective
Medium, Blaser-Wang
(Blaser-Wang Campylobacter Medium)**

Composition per liter:

Columbia agar base.....	890.0mL
Sheep blood	100.0mL
Antibiotic supplement.....	10.0mL
pH 7.3 ± 0.2 at 25°C	

Columbia Agar Base:**Composition per liter:**

Special peptone.....	25.0g
Agar	10.0g
NaCl.....	5.0g
Starch	1.0g

Preparation of Columbia Agar Base: Add components to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:**Composition per 10.0mL:**

Cephalothin.....	15.0mg

Vancomycin	10.0mg
Trimethoprim	5.0mg
Amphotericin B	2.0mg
Polymyxin B	2,500U

Preparation of Antibiotic Supplement: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: To 890.0mL of cooled, sterile Columbia agar base, aseptically add 100.0mL of sheep blood or 50.0–70.0mL of laked horse blood. Laked blood is prepared by freezing whole blood overnight and thawing to room temperature. Aseptically add 10.0mL of sterile antibiotic supplement. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species.

Campylobacter Selective Medium, Butzler's

(Butzler's *Campylobacter* Medium)

Composition per liter:

<i>Brucella</i> agar base	940.0mL
Sheep or horse blood, defibrinated	50.0mL
Antibiotic supplement	10.0mL
pH 7.5 ± 0.2 at 25°C	

***Brucella* Agar Base:**

Composition per liter:

Agar	15.0g
Glucose	10.0g
Pancreatic digest of casein	10.0g
NaCl	5.0g
Peptic digest of animal tissue	5.0g

Preparation of *Brucella* Agar Base: Add components to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:

Composition per 10.0mL:

Cycloheximide	50.0mg
Cephazolin	15.0mg
Novobiocin	5.0mg
Bacitracin	25,000U
Colistin sulfate	10,000U

Preparation of Antibiotic Supplement: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: To 940.0mL of cooled, sterile *Brucella* agar base, aseptically add 50.0mL of defibrinated sheep or horse blood and 10.0mL of sterile antibiotic supplement. Mix thoroughly. For

enhanced growth, medium may also be supplemented with 0.25g of Fe₂SO₄·H₂O, 0.25g of sodium metabisulfite, and 0.25g of sodium pyruvate. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species.

Campylobacter Selective Medium, Butzler's

(Butzler's *Campylobacter* Medium)

Composition per liter:

Columbia agar base	940.0mL
Blood, horse or sheep	50.0mL
Antibiotic supplement	10.0mL
pH 7.3 ± 0.2 at 25°C	

Columbia Agar Base:

Composition per liter:

Peptone	25.0g
Agar	10.0g
NaCl	5.0g
Starch	1.0g

Preparation of Columbia Agar Base: Add components to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:

Composition per 10.0mL:

Cycloheximide	50.0mg
Cephazolin	15.0mg
Novobiocin	5.0mg
Bacitracin	25,000U
Colistin sulfate	10,000U

Preparation of Antibiotic Supplement: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: To 940.0mL of cooled, sterile Columbia agar base, aseptically add 50.0mL of defibrinated sheep or horse blood and 10.0mL of sterile antibiotic supplement. Mix thoroughly. The medium may also be supplemented with 0.25g of Fe₂SO₄·H₂O, 0.25g of sodium metabisulfite, and 0.25g of sodium pyruvate. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species.

Campylobacter Selective Medium, Karmali's

(Karmali's *Campylobacter* Medium)

Composition per liter:

Activated charcoal	4.0g

Columbia agar base.....	990.0mL
Antibiotic supplement.....	10.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Columbia Agar Base:

Composition per 990.0mL:

Peptone.....	25.0g
Agar.....	10.0g
NaCl.....	5.0g
Starch.....	1.0g

Preparation of Columbia Agar Base: Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:

Composition per 10.0mL:

Sodium pyruvate.....	0.05g
Cycloheximide.....	0.05g
Cefoperazone.....	0.016g
Hemin.....	0.016g
Vancomycin.....	0.01g

Preparation of Antibiotic Supplement: Add components to 10.0mL of distilled/deionized water. Filter sterilize.

Preparation of Medium: Prepare 990.0mL of Columbia agar base. Sterilize as directed. Cool to 50°–55°C. Add defibrinated sheep or horse blood to a final concentration of 5–7%. Add 10.0mL of sterile antibiotic supplement. Mix thoroughly. For enhanced growth, medium may also be supplemented with 0.25g of Fe₂SO₄·H₂O, 0.25g of sodium metabisulfite, and 0.25g of sodium pyruvate. Pour into sterile Petri dishes. Swirl while pouring to keep charcoal in suspension.

Use: For the selective isolation of *Campylobacter* species.

***Campylobacter* Selective Medium, Preston's**

(Preston's *Campylobacter* Medium)

Composition per liter:

<i>Campylobacter</i> agar base.....	940.0mL
Horse blood, lysed.....	50.0mL
Antibiotic supplement.....	10.0mL
pH 7.5 ± 0.2 at 25°C	

***Campylobacter* Agar Base:**

Composition per liter:

Agar.....	12.0g
Beef extract.....	10.0g

Peptone.....	10.0g
NaCl.....	5.0g

Preparation of *Campylobacter* Agar Base:

Add components to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Supplement:

Composition per 10.0mL:

Cycloheximide.....	0.1g
Rifampicin.....	0.01g
Trimethoprim lactate.....	0.01g
Polymyxin B.....	5000U

Preparation of Antibiotic Supplement: Add components to 10.0mL of 50:50 acetone:distilled/deionized water. Filter sterilize.

Preparation of Medium: To 940.0mL of cooled, sterile *Campylobacter* agar base, aseptically add 50.0mL of lysed horse blood and 10.0mL of sterile antibiotic supplement. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the selective isolation of *Campylobacter* species.

Carbohydrate Fermentation Broth

Composition per liter:

Peptone.....	10.0g
NaCl.....	5.0g
Meat extract.....	3.0g
Carbohydrate solution.....	50.0mL
Andrade's indicator.....	10.0mL
pH 7.1 ± 0.2 at 25°C	

Andrade's Indicator:

Composition per 100.0mL:

Acid Fuchsin.....	0.1g
NaOH (1N solution).....	16.0mL

Preparation of Andrade's Indicator: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Adonitol, arabinose, cellobiose, glucose, dulcitol, fructose, galactose, inositol, lactose, maltose, mannitol, raffinose, rhamnose, salicin, sorbitol, sucrose, trehalose, xylose, or other carbohydrates may be used. Mix thoroughly. Filter sterilize.

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute in 10.0mL volumes into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 25°C. Add 0.5mL of sterile carbohydrate solution to each tube.

Use: For the determination of carbohydrate fermentation reactions of microorganisms, particularly members of the Enterobacteriaceae. A Durham tube is used to collect gas produced during the fermentation reaction. Acid production is indicated by a pink reaction.

Casamino Acids Yeast Extract Broth (CYE Broth)

Composition per liter:

Casamino acids	30.0g
Yeast extract	4.0g
K ₂ HPO ₄	0.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure—121°C.

Use: For the cultivation of *Vibrio* species from foods.

Cary and Blair Transport Medium (BAM M31)

Composition per liter:

Agar	5.0g
NaCl	5.0g
Sodium thioglycolate	1.5g
Na ₂ HPO ₄	1.1g
CaCl ₂ solution	9.0mL
pH 8.4 ± 0.5 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

CaCl₂ Solution:

Composition per 10.0mL:

CaCl ₂	0.1g

Preparation of CaCl₂ Solution: Add CaCl₂ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except CaCl₂ solution, to distilled/deionized water and bring volume to 991.0mL. Mix thoroughly and heat

gently until boiling. Cool to 50°C. Add 9.0mL of a 1% CaCl₂ solution. Adjust the pH to 8.4. Distribute into screw-capped tubes in 7.0mL volumes. Sterilize under flowing steam for 15 min. After sterilization, tighten the screwcaps.

Use: For the maintenance—as a holding medium or transport medium—of specimens during collection or shipment.

Casamino Acids Yeast Extract Broth (CYE Broth) (BAM M32)

Composition per liter:

Casamino acids	30.0g
Yeast extract	4.0g
K ₂ HPO ₄	0.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure—121°C.

Use: For the cultivation of *Vibrio* species from foods.

Casamino Acids Yeast Extract Lincomycin Medium

Composition per liter:

Casamino acids	20.0g
K ₂ HPO ₄	8.71g
Yeast extract	6.0g
NaCl	2.5g
Lincomycin solution	5.0mL
Trace salts solution	1.0mL
pH 8.5 ± 0.2 at 25°C	

Lincomycin Solution:

Composition per 5.0mL:

Lincomycin	45.0mg

Preparation of Lincomycin Solution: Add lincomycin to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Trace Salts Solution:

Composition per liter:

MgSO ₄ ·7H ₂ O	50.0g
MnCl ₂ ·4H ₂ O	5.0g
FeCl ₂	5.0g

Preparation of Trace Salts Solution: Add components to distilled/deionized water and bring volume to 1.0L. Add sufficient 0.1N H₂SO₄ to dissolve components. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except trace salts solution and lincomycin solution, to distilled/deionized water and bring volume to

994.0mL. Mix thoroughly. Adjust pH to 8.5. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 1.0mL of sterile trace salts solution and 5.0mL of sterile lincomycin solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of heat-labile, toxin-producing enterotoxigenic *Escherichia coli*.

Casamino Acids Yeast Extract Salts Broth, Gorbach (CA YE Broth) (BAM M34)

Composition per liter:

Casamino acids	20.0g
K ₂ HPO ₄	8.71g
Yeast extract	6.0g
NaCl	2.5g
Trace salts solution	1.0mL
pH 8.5 ± 0.2 at 25°C	

Trace Salts Solution:

Composition per liter:

MgSO ₄ ·7H ₂ O	50.0g
MnCl ₂ ·4H ₂ O	5.0g
FeCl ₂	5.0g

Preparation of Trace Salts Solution: Add components to distilled/deionized water and bring volume to 1.0L. Add sufficient 0.1N H₂SO₄ to dissolve components. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except trace salts solution, to distilled/deionized water and bring volume to 999.0mL. Mix thoroughly. Adjust pH to 8.5. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 1.0mL of sterile trace salts solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of enterotoxigenic *Escherichia coli*.

CCY Modified Medium

Composition per liter:

Yeast extract	30.0g
Casamino acids	20.0g
Na ₂ HPO ₄	2.48g
KH ₂ PO ₄	0.41g
MgSO ₄ ·7H ₂ O	20.0mg
MnSO ₄ ·H ₂ O	7.5mg
Citric acid	6.4mg
FeSO ₄ ·7H ₂ O	6.4mg
Sodium pyruvate solution	100.00mL
pH 7.3 ± 0.2 at 25°C	

Sodium Pyruvate Solution:

Composition per 100.0mL:

Sodium pyruvate	23.2g

Preparation of Sodium Pyruvate Solution:

Add sodium pyruvate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sodium pyruvate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.3. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 100.0mL of sterile sodium pyruvate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Staphylococcus aureus*.

Cefsulodin Irgasan Novobiocin Agar (CIN Agar) (Yersinia Selective Agar) (BAM M35)

Composition per 1008.0mL:

Basal medium	757.0mL
Deoxycholate solution	200.0mL
Cefsulodin solution	10.0mL
Novobiocin solution	10.0mL
Crystal Violet solution	10.0mL
Strontium chloride solution	10.0mL
Neutral Red solution	10.0mL
NaOH, 5N	1.0mL
Irgasan solution	1.0mL
pH 7.4 ± 0.2 at 25°C	

Basal Medium:

Composition per 757.0mL:

Mannitol	20.0g
Special peptone	20.0g
Agar	12.0g
Sodium pyruvate	2.0g
Yeast extract	2.0g
NaCl	1.0g
Magnesium sulfate solution	1.0mL

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 757.0mL. Mix thoroughly. Gently heat and bring to boiling with stirring. Cool to about 80°C by placing in a 50°C water bath for about 10 min.

Magnesium Sulfate Solution:

Composition per 10mL:

MgSO ₄ ·7H ₂ O	0.1g

Preparation of Magnesium Sulfate Solution:

Add MgSO₄·7H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Irgasan Solution:**Composition** per 10mL:

Irgasan (triclosan).....0.04g

Preparation of Irgasan Solution: Add irgasan to 95% ethanol and bring volume to 10.0mL. Mix thoroughly. Can be stored for 4 weeks at -20°C.

Deoxycholate Solution:**Composition** per 200.0mL:

Na-deoxycholate 0.5g

Preparation of Deoxycholate Solution: Add deoxycholate to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Gently heat and bring to boiling with stirring. Cool to 50–55°C.

Neutral Red Solution:**Composition** per 10.0mL:

Neutral Red 30.0mg

Preparation of Neutral Red Solution: Add Neutral Red to 10.0mL of distilled/deionized water. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Crystal Violet Solution:**Composition** per 10.0mL:

Crystal Violet 1.0mg

Preparation of Crystal Violet Solution: Add Crystal Violet to 10.0mL of distilled/deionized water. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Cefsulodin Solution:**Composition** per 10.0mL:

Cefsulodin 15.0mg

Preparation of Cefsulodin Solution: Add cefsulodin to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Novobiocin Solution:**Composition** per 10.0mL:

Novobiocin 2.5mg

Preparation of Novobiocin Solution: Add novobiocin to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Strontium Chloride Solution:**Composition** per 10.0mL:SrCl₂·6H₂O 1.0g

Preparation of Strontium Chloride: Add strontium chloride to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add 1.0mL irgasan solution to 757.0mL basal medium. Mix thoroughly. Cool to 50–55°C. Add 200.0mL desoxycholate solution. Mix thoroughly. Solution should remain clear.

Aseptically add 1.0mL 5*N* NaOH, 10.0mL Neutral Red solution, 10.0mL Crystal Violet solution, 10.0mL cefsulodin solution, and 10.0mL novobiocin solution. Mix thoroughly. Slowly add 10.0mL strontium chloride solution while continuously stirring. Adjust pH to 7.4 with 5*N* NaOH. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and differentiation of *Yersinia enterocolitica* based on mannitol fermentation. *Yersinia enterocolitica* appears as “bull’s eye” colonies with deep red centers surrounded by a transparent periphery.

Cell Growth Medium (BAM M36)

Composition per 2250.0mL:

Eagle’s minimal essential medium

with Hanks’ salts (MEMH) 1.0L

L 15 medium, modified Leibovitz 1.0L

Fetal calf serum 200.0mL

NaHCO₃ solution 50.0mL

pH 7.5 at 25°C

MEMH:**Composition** per liter:

NaCl 8.0g

Glucose 1.0g

KCl 0.4g

CaCl₂·2H₂O 0.14gMgSO₄·7H₂O 0.1gKH₂PO₄ 0.06gNa₂HPO₄ 0.05g

L-Isoleucine 0.026g

L-Leucine 0.026g

L-Lysine 0.026g

L-Threonine 0.024g

L-Valine 0.0235g

L-Tyrosine 0.018g

L-Arginine 0.0174g

L-Phenylalanine 0.0165g

L-Cystine 0.012g

L-Histidine 8.0mg

L-Methionine 7.5mg

Phenol Red 5.0mg

L-Tryptophan 4.0mg

Inositol 1.8mg

Biotin 1.0mg

Folic acid 1.0mg

Calcium pantothenate 1.0mg

Choline chloride 1.0mg

Nicotinamide 1.0mg

Pyridoxal·HCl 1.0mg

Thiamine·HCl 1.0mg

Riboflavin 0.1mg

Preparation of MEMH: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

L 15 Medium, Modified Leibovitz

Composition per liter:

NaCl.....	8.0g
DL-Threonine.....	0.6g
Sodium pyruvate.....	0.6g
DL-Alanine.....	0.5g
L-Arginine, free base.....	0.5g
KCl.....	0.4g
L-Asparagine·H ₂ O.....	0.3g
L-Histidine, free base.....	0.3g
L-Glutamine.....	0.3g
L-Isoleucine.....	0.3g
L-Phenylalanine.....	0.3g
L-Tyrosine.....	0.3g
DL-Methionine.....	0.2g
DL-Valine.....	0.2g
Glycine.....	0.2g
L-Serine.....	0.2g
Na ₂ HPO ₄ , anhydrous.....	0.2g
CaCl ₂ , anhydrous.....	0.1g
L-Cysteine, free base.....	0.1g
L-Leucine·HCl.....	0.1g
Streptomycin.....	0.1g
MgCl ₂ , anhydrous.....	0.094g
D-Galactose.....	0.09g
KH ₂ PO ₄	0.06g
Gentamicin.....	0.05g
L-Tryptophan.....	0.02g
Phenol Red.....	0.01g
<i>i</i> -Inositol.....	2.0mg
Choline chloride.....	1.0mg
D-Calcium pantothenate.....	1.0mg
Folic acid.....	1.0mg
Nicotinamide.....	1.0mg
Pyridoxine·HCl.....	1.0mg
Thiamine monophosphate·2H ₂ O.....	1.0mg
Riboflavin-5-phosphate.....	0.1mg
Penicillin G.....	100,000U

pH 7.5 ± 0.2 at 25°C

Preparation of L 15 Medium, Modified Leibovitz: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Store at 5°C.

NaHCO₃ Solution:

Composition per 100.0mL:

NaHCO ₃	7.5g

Preparation of NaHCO₃ Solution: Add NaHCO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine 1.0L of sterile Eagle's minimal essential medium with Hanks' salts (MEMH) and 1.0L of sterile L 15 medium, modified Leibovitz. Mix thoroughly. Immediately prior to use, aseptically add 200.0mL of fetal calf serum and 50.0mL of NaHCO₃ solution. Mix thoroughly. Aseptically distribute into sterile containers.

Use: For the cultivation of mammalian HeLa or Vero tissue culture cells to test the cytopathic effects of *Escherichia coli*.

Cellobiose Polymyxin Colistin Agar (CPC Agar)

Composition per liter:

Solution A.....	900.0mL
Solution B.....	100.0mL

pH 7.6 ± 0.2 at 25°C

Solution A:

Composition per 900.0mL:

NaCl.....	20.0g
Agar.....	15.0g
Peptone.....	10.0g
Beef extract.....	5.0g
Bromthymol Blue.....	0.04g
Cresol Red.....	0.04g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.6. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution B:

Composition per 100.0mL:

Cellobiose.....	15.0g
Colistin.....	1,360,000U
Polymyxin B.....	100,000U

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine 900.0mL of cooled, sterile solution A and 100.0mL of sterile solution B. Mix thoroughly. Pour into sterile Petri dishes. Use within 7 days.

Use: For the cultivation and identification of *Vibrio* species from foods.

Cellobiose Polymyxin B Colistin Agar, Modified (mCPC Agar) (BAM M98)

Composition per liter:

Solution 1.....	900.0mL

Solution 2 100.0mL
pH 7.6 ± 0.2 at 25°C

Solution 1:**Composition** per 900.0mL:

NaCl 20.0g
Agar 15.0g
Peptone 10.0g
Beef extract 5.0g
1000× dye stock solution 1.0mL

Preparation of Solution 1: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.6. Gently heat and bring to boiling. Do not autoclave. Cool to 48° – 55°C .

1000X Dye Stock Solution:**Composition** per 100.0mL:

Bromthymol Blue 4.0g
Cresol Red 4.0g
Ethanol (95% solution) 100.0mL

Preparation of 1000X Dye Stock Solution: Add Bromthymol Blue and Cresol Red to 100.0mL of ethanol. Mix thoroughly.

Solution 2:**Composition** per 100.0mL:

Cellobiose 10.0g
Colistin 400,000U
Polymyxin B 100,000U

Preparation of Solution 2: Add cellobiose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat until dissolved. Cool to 25°C . Add colistin and polymyxin B. Mix thoroughly.

Preparation of Medium: Combine cooled solution 1 and solution 2. Mix thoroughly. Do not autoclave. Pour into sterile Petri dishes.

Use: For the cultivation of *Vibrio* species from foods.

Cellulase Solution (BAM M187)

Composition per 100.0mL:

Cellulase 1.0g

Preparation of Medium: Add cellulase to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. filter sterilize.

Use: For the pre-treatment of guar gum in the analysis of *Salmonella* spp. The cellulase reduces the viscosity of the guar gum so that it can be pipetted into broth or agar media.

Cereal Agar

Composition per liter:

Cereal, precooked mixed 100.0g
Agar 15.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation and sporulation of fungi.

Cetrimide Agar, Non-USP

Composition per liter:

Beef heart, solids from infusion 500.0g
Agar 15.0g
Tryptose 10.0g
NaCl 5.0g
Cetrimide 0.9g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure– 118°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and identification of *Pseudomonas aeruginosa* and other Gram-negative, nonfermentative bacteria.

Cetrimide Agar, USP (Pseudosel® Agar) (BAM M37)

Composition per liter:

Pancreatic digest of gelatin 20.0g
Agar 13.6g
 K_2SO_4 10.0g
 MgCl_2 1.4g
Cetrimide 0.3g
Glycerol 10.0mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure– 118°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and identification of *Pseudomonas aeruginosa* and other Gram-negative, nonfermentative bacteria.

Chapman Stone Agar

Composition per liter:

(NH ₄) ₂ SO ₄	75.0g
NaCl.....	55.0g
Gelatin.....	30.0g
Agar	15.0g
D-Mannitol	10.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	5.0g
Yeast extract.....	2.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes while the medium is still hot. Add 25.0mL of medium per Petri dish.

Use: For the isolation of staphylococci from a variety of specimens.

Cheese Agar

Composition per liter:

Cheese, ripened.....	100.0g
NaCl.....	50.0g
Agar	15.0g
Peptone.....	10.0g
Potassium citrate.....	10.0g
Sodium oxalate	2.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add the cheese and potassium citrate to distilled/deionized water and bring volume to 300.0mL. Gently heat and bring to 50°C to separate the fat. Discard the fat. In a separate flask, add the remaining components to distilled/deionized water and bring volume to 700.0mL. Gently heat and bring to boiling. Add the 300.0mL of aqueous suspension of cheese solids. Adjust pH to 7.4. Autoclave for 25 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Brevibacterium linens*.

Cherry Agar, CBS Formula

Composition per liter:

Pulp of sour stone cherries.....	200.0g

Agar	20.0g

pH 3.8 ± 0.2 at 25°C

Preparation of Medium: Add 1.0L of distilled/deionized water to pulp of sour stone cherries. Gently heat and bring to boiling. Simmer gently for 2 hr. Strain through cloth. Autoclave filtrate for 30 min at 6 psi pressure–110°C. Add agar to distilled/deionized water and bring volume to 800.0mL. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. To sterile agar solution, add 200.0mL of sterile cherry filtrate. Distribute into sterile tubes. Autoclave for 5 min at 15 psi pressure–121°C.

Use: For the cultivation of various fungi.

China Blue Lactose Agar

Composition per liter:

Agar	12.0g
Lactose.....	10.0g
Peptone	5.0g
NaCl.....	5.0g
Beef extract.....	3.0g
China Blue	0.375g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation, differentiation, and enumeration of bacteria from dairy products. Lactose-fermenting bacteria appear as blue colonies. Nonlactose-fermenting bacteria appear as colorless colonies.

CHO Medium Base (Carbohydrate Medium Base)

Composition per liter:

Pancreatic digest of casein.....	15.0g
Yeast extract.....	7.0g
NaCl.....	2.5g
Agar	0.75g
Sodium thioglycolate	0.5g
L-Cystine	0.25g
Ascorbic acid	0.1g
Bromthymol Blue	0.01g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Use: Used as a basal medium to which carbohydrates are added for fermentation studies of anaerobic bacteria. Generally, 6.25mL of a 10% filter-sterilized solution of carbohydrate is added to the sterile basal medium.

Cholera Medium TCBS

Composition per liter:

Sucrose.....	20.0g
Agar	14.0g
Peptone.....	10.0g
NaCl.....	10.0g
Sodium citrate.....	10.0g
Na ₂ S ₂ O ₃ ·5H ₂ O.....	10.0g
Ox bile.....	8.0g
Yeast extract.....	5.0g
Ferric citrate.....	1.0g
Bromthymol Blue	0.04g
Thymol Blue	0.04g

pH 8.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Pour into sterile Petri dishes. Dry agar plates before using.

Use: For the growth of *Vibrio cholerae*, *Vibrio parahaemolyticus*, and other *Vibrio* species.

Chopped Liver Broth (BAM M38)

Composition per liter:

Fresh beef liver	500.0g
Peptone.....	10.0g
K ₂ HPO ₄	1.0g
Soluble starch.....	1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Grind fresh beef liver. Add to 1.0L of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 60 min. Cool to 25°C. Adjust pH to 7.0. Gently heat and bring to boiling. Continue boiling for 10 min. Filter through cheesecloth. Press out excess liquid. Save chopped liver particles. To filtrate, add remaining components. Bring volume to 1.0L with distilled/deionized water. Adjust pH to 7.0. Filter through Whatman #1 filter paper. Add chopped liver particles to test tubes to a depth of 1.2–2.5 cm. Add 10.0mL of

broth to each tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Clostridium botulinum*, *Clostridium perfringens*, and other anaerobic bacteria from foods.

Chopped Meat Agar

Composition per liter:

Ground meat, fat free.....	500.0g
Pancreatic digest of casein.....	30.0g
Agar	15.0g
K ₂ HPO ₄	5.0g
Yeast extract.....	5.0g
L-Cysteine-HCl	0.5g
Resazurin	1.0mg
NaOH (1N solution).....	25.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Use lean beef or horse meat. Remove fat and connective tissue. Grind finely. Add ground meat and 25.0mL of NaOH solution to distilled/deionized water and bring volume to 1025mL. Gently heat and bring to boiling. Continue boiling for 15 min without stirring. Cool to room temperature. Remove fat from surface. Filter and retain both meat particles and filtrate. Adjust volume of filtrate to 1.0L with distilled/deionized water. Add pancreatic digest of casein, agar, K₂HPO₄, yeast extract, and resazurin. Gently heat and bring to boiling. Boil for 1–2 min. Add L-cysteine-HCl. Mix thoroughly. Distribute 7.0mL into tubes that contain meat particles (1 part meat particles to 5 parts fluid). Autoclave for 30 min at 15 psi pressure–121°C.

Use: For the cultivation of various anaerobes.

Chopped Meat Broth

Composition per liter:

Ground meat, fat free.....	500.0g
Pancreatic digest of casein.....	30.0g
K ₂ HPO ₄	5.0g
Yeast extract.....	5.0g
L-Cysteine-HCl	0.5g
Resazurin	1.0mg
NaOH (1N solution).....	25.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Use lean beef or horse meat. Remove fat and connective tissue. Grind finely. Add ground meat and 25.0mL of NaOH solution to distilled/deionized water and bring volume to 1025mL. Gently heat and bring to boiling. Continue boiling for 15 min without stirring. Cool to room temperature. Remove fat from surface. Filter and retain both meat particles and filtrate. Adjust volume of filtrate to 1.0L with distilled/deionized water. Add

pancreatic digest of casein, K_2HPO_4 , yeast extract, and resazurin. Gently heat and bring to boiling. Boil for 1–2 min. Add L-cysteine-HCl. Mix thoroughly. Distribute 7.0mL into tubes that contain meat particles (1 part meat particles to 5 parts fluid). Autoclave for 30 min at 15 psi pressure–121°C.

Use: For the cultivation of various anaerobes.

Chopped Meat Carbohydrate Medium

Composition per 1240.0mL:

Peptone.....	30.0g
K_2HPO_4	5.0g
Yeast extract.....	5.0g
Cellobiose.....	1.0g
Maltose.....	1.0g
Starch.....	1.0g
L-Cysteine-HCl·H ₂ O.....	0.5g
Chopped meat extract filtrate.....	1.0L
Chopped meat extract solids.....	200.0mL
Resazurin (0.025% solution).....	4.0mL
pH 7.0 ± 0.2 at 25°C	

Chopped Meat Extract:

Composition per liter:

Beef or horse meat.....	500.0g
NaOH (1N solution).....	25.0mL

Preparation of Chopped Meat Extract: Use lean beef or horse meat. Remove fat and connective tissue. Grind. Add meat and NaOH to distilled/deionized water and bring volume to 1.0L. Gently heat and bring to boiling while stirring. Cool to 25°C. Remove fat from surface. Filter. Reserve ground meat particles and filtrate. Add distilled/deionized water to filtrate and bring volume to 1.0L.

Preparation of Medium: To 1.0L of chopped meat extract filtrate, add the remaining components, except the L-cysteine-HCl·H₂O and chopped meat solids. Mix thoroughly. Gently heat to boiling. Cool to room temperature. Add the L-cysteine-HCl·H₂O. Adjust pH to 7.0. Distribute 1 part chopped meat solids (by volume) and 5 parts of liquid (by volume) into tubes under O₂-free 97% N₂ + 3% H₂. Cap with rubber stoppers and place tubes in a press. Autoclave for 15 min at 15 psi pressure–121°C with fast exhaust.

Use: For the cultivation of anaerobic bacteria, including *Clostridium* species.

Christensen Agar

Composition per liter:

Agar.....	15.0g
NaCl.....	5.0g
Sodium citrate.....	3.0g

KH_2PO_4	1.0g
L-Cysteine-HCl·H ₂ O.....	0.1g
Phenol Red.....	12.0mg
pH 6.9 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation of enteric pathogens, especially members of the Enterobacteriaceae, and coliforms based on their ability to utilize citrate as a carbon source. Bacteria that can utilize citrate turn the medium pink-red.

Christensen Agar

Composition per liter:

Agar.....	15.0g
NaCl.....	5.0g
Sodium citrate.....	3.0g
KH_2PO_4	1.0g
Yeast extract.....	0.5g
Glucose.....	0.2g
L-Cysteine-HCl·H ₂ O.....	0.1g
Phenol Red.....	12.0mg
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation of enteric pathogens, especially members of the Enterobacteriaceae, and coliforms based on their ability to utilize citrate as a carbon source. Bacteria that can utilize citrate turn the medium pink-red.

Christensen Citrate Agar (BAM M39)

Composition per liter:

Agar.....	15.0g
NaCl.....	5.0g
Sodium citrate.....	3.0g
KH_2PO_4	1.0g
Yeast extract.....	0.5g
Ferric ammonium citrate.....	0.4g
L-Cysteine-HCl·H ₂ O.....	0.1g
Na ₂ S ₂ O ₅	0.08g

Phenol Red 12.0mg
pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation of enteric pathogens, especially members of the Enterobacteriaceae, and coliforms based on their ability to utilize citrate as a carbon source. Bacteria that can utilize citrate turn the medium pink-red.

Christensen Citrate Agar, Modified (Citrate Agar)

Composition per liter:

Agar 12.0g
NaCl 5.0g
Sodium citrate 3.8g
 KH_2PO_4 1.0g
Yeast extract 0.5g
Glucose 0.2g
L-Cysteine·HCl· H_2O 0.1g
Phenol Red 0.02g
pH 6.7 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation of enteric pathogens, especially members of the Enterobacteriaceae, and coliforms based on their ability to utilize citrate as a carbon source. Bacteria that can utilize citrate turn the medium pink-red.

Christensen Citrate Sulfide Medium

Composition per liter:

Agar 15.0g
NaCl 5.0g
Sodium citrate· $2\text{H}_2\text{O}$ 3.0g
 KH_2HPO_4 1.0g
Yeast extract 0.5g
Ferric citrate 0.2g
Ammonium citrate 0.2g
Glucose 0.2g
L-Cysteine·HCl· H_2O 0.1g
 $\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$ 0.08g
Phenol Red 0.012g
pH 6.7 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation of enteric pathogens, especially members of the Enterobacteriaceae, and coliforms based on their ability to utilize citrate as a carbon source and production of H_2S . Bacteria that can utilize citrate turn the medium pink-red. H_2S production appears as a blackening of the butt of the tube.

Christensen's Urea Agar (BAM M40)

Composition per liter:

Agar 15.0g
NaCl 5.0g
 KH_2PO_4 2.0g
Peptone 1.0g
Glucose 1.0g
Phenol Red 0.012g
Urea solution 100.0mL
pH 6.8 ± 0.1 at 25°C

Urea Solution:

Composition per 100.0mL:

Urea 20.0g

Preparation of Urea Solution: Add urea to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except urea solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 50 – 55°C . Aseptically add 100.0mL of sterile urea solution. Mix thoroughly. Pour into Petri dishes or distribute into sterile tubes. Allow tubes to solidify in a slanted position.

Use: For the differentiation of a variety of microorganisms, especially members of the Enterobacteriaceae, aerobic actinomycetes, streptococci, and nonfermenting Gram-negative bacteria, on the basis of urease production.

Christensen's Urea Agar with NaCl (BAM M40)

Composition per liter:

NaCl 20.0g
Agar 15.0g
 KH_2PO_4 2.0g
Peptone 1.0g
Glucose 1.0g

Phenol Red.....	0.012g
Urea solution.....	100.0mL
pH 6.8 ± 0.1 at 25°C	

Urea Solution:**Composition per 100.0mL:**

Urea.....	20.0g

Preparation of Urea Solution: Add urea to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except urea solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50–55°C. Aseptically add 100.0mL of sterile urea solution. Mix thoroughly. Pur into Petri dishes or distribute into sterile tubes. Allow tubes to solidify in a slanted position.

Use: For the differentiation of halophilic *Vibrio* spp. on the basis of urease production.

CHROMagar *E. coli***Composition per liter:**

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding tellurite can increase specificity. Cool to 45–50°C. Pour into sterile Petri dishes.

Source: CHROMagar *E. coli* is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Escherichia coli* which forms blue colonies.

CHROMagar ECC**Composition per liter:**

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding tellurite can increase specificity. Cool to 45–50°C. Pour into sterile Petri dishes.

Source: CHROMagar EEC is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Escherichia coli* and other coliform bacteria which form red colonies.

**CHROMagar *Listeria*
(BAM M40a)****Composition per liter:**

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding tellurite can increase specificity. Cool to 45–50°C. Pour into sterile Petri dishes.

Source: CHROMagar *Listeria* is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Listeria monocytogenes* which form blue colonies surrounded by white halos.

CHROMagar O157**Composition per liter:**

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding tellurite can increase specificity. Cool to 45–50°C. Pour into sterile Petri dishes.

Source: CHROMagar O157 is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Escherichia coli* O157.

CHROMagar *Salmonella***Composition per liter:**

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Cool to 45–50°C. Pour into sterile Petri dishes.

Source: CHROMagar *Salmonella* is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Salmonella* spp.

CHROMagar *Staph. aureus*

Composition per liter:

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding an antibiotic such as tobramycin or methicillin can be used to identify resistant strains. Cool to 45-50°C. Pour into sterile Petri dishes.

Source: CHROMagar *Staph. aureus* is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Staphylococcus aureus*.

CHROMagar *Vibrio*

Composition per liter:

Proprietary

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath or steam bath. Shake periodically during heating to dissolve components. Heat long enough with shaking every 5 min to ensure complete dissolution. Do not overheat. Adding tellurite can increase specificity. Cool to 45-50°C. Pour into sterile Petri dishes.

Source: CHROMagar *Vibrio* is available from CHROMagar Microbiology.

Use: For the differentiation and presumptive identification of *Vibrio parahaemolyticus* which form mauve colonies; *Vibrio cholerae* form turquoise blue colonies, and *Vibrio alginolyticus* colonies are colorless.

Chromogenic *Bacillus cereus* Agar

Composition per liter:

Agar	13.0g
Peptone.....	10.0g
Sodium pyruvate.....	10.0g
Yeast extract.....	4.0g
Na ₂ HPO ₄	2.52
Chromogenic mix	1.2g
KH ₂ PO ₄	0.28g
Selective supplement solution	10.0mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Selective Supplement Solution:

Composition per 10.0mL:

Trimethoprim	10.0mg
Polymyxin B sulfate	106,000U

Preparation of Selective Supplement Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except selective supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL selective supplement solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: A chromogenic medium for the isolation and differentiation of *Bacillus cereus* from food samples.

Chromogenic *E. coli*/Coliform Medium

Composition per liter:

Chromogenic mix	20.3g
Agar	15.0g
Peptone	5.0g
NaCl.....	5.0g
Na ₂ HPO ₄	3.5g
Yeast extract.....	3.0g
Lactose.....	2.5g
NaH ₂ PO ₄	1.5g
Neutral Red.....	0.03g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation between *Escherichia coli* and other coliforms in cultures produced from food samples. Agar base uses two enzyme substrates to differentiate between *E. coli* and other coliforms. One chromogenic substrate is cleaved by the enzyme glucuronidase which is specific for *E. coli* and produced by approximately 97% of strains. The second chromogenic substrate is cleaved by galactosidase, an enzyme produced by the majority of coliforms. This results in purple *E. coli* colonies, as they are able to cleave both chromogenic substrates and pink coliform colonies as they are only able to cleave the galactosidase chromogen.

Chromogenic *Enterobacter sakazakii***Agar, DFI Formulation****Composition per liter:**

Agar	15.0g
Tryptone	15.0g
Soya peptone	5.0g
NaCl	5.0g
Ferric ammonium citrate	1.0g
Sodium deoxycholate	1.0g
Na ₂ S ₂ O ₃	1.0g
Chromogen	0.1g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Dispense into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the differentiation and enumeration of *Enterobacter sakazakii* from infant formula and other food samples. The enzyme α -glucosidase, present in *E. sakazakii*, hydrolyzes the substrate 5-bromo-4-chloro-3-indolyl- α ,D-glucopyranoside, thus producing blue-green colonies on this pale yellow medium. *Proteus vulgaris* is also weakly α -glucosidase positive and could grow to give colonies of a similar color to *E. sakazakii*. However, on this medium, *Proteus* spp. grow as grey colonies: they produce hydrogen sulphide in the presence of ferric ions forming ferrous sulphide. Deoxycholate inhibits the growth of most Gram-positive organisms.

**Chromogenic *Listeria* Agar
(BAM M40b)****Composition per liter:**

Proprietary

Source: This medium is available as a premixed powder from Oxoid.

Use: For the isolation, enumeration and presumptive identification of *Listeria* spp. and *Listeria monocytogenes* from food samples. This selective medium contains the substrate lecithin, which permits differentiation of *L. monocytogenes* and *L. ivanovii* from other *Listeria* species. Differential activity for all *Listeria* species is due to the addition of a chromogenic substrate.

**Chromogenic *Salmonella* Esterase Agar
(CSE Agar)****Composition per liter:**

Lactose	14.65
Agar	12.0g
Peptone	4.0g
Tryptone	4.0g

Tween 20	3.0g
Lab Lemco	3.0g
Na ₃ -citrate dihydrate	0.5g
L-cysteine	0.128g
Tris	0.06g
SLA-octonoate solution	50.0mL
Novobiocin solution	10.0mL
Ethyl 4-dimethylaminobenzoate solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Novobiocin Solution:**Composition per 10mL:**

Novobiocin	70.0mg

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Ethyl 4-dimethylaminobenzoate Solution:**Composition per 10mL:**

Ethyl 4-dimethylaminobenzoate	0.35g
Methanol	8.0mL

Preparation of Ethyl 4-dimethylaminobenzoate Solution: Add ethyl 4-dimethylaminobenzoate to 8.0mL methanol. Mix thoroughly. Bring volume to 10.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

SLA-Octonoate Solution:**Composition per 50mL:**

4-[2-(4-octanoyloxy-3,5-dimethoxyphenyl)-vinyl]-quinolinium-1-(propan-3-yl carboxylic acid) bromide (SLPA-octanoate; bromide form)	0.3223g

Preparation of SLA-Octonoate Solution: Add SLA-octonoate to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, SLA-octonoate solution, and ethyl 4-dimethylaminobenzoate solution, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL novobiocin solution, 50.0mL SLA-octonoate solution, and 10.0mL ethyl 4-dimethylaminobenzoate solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the differentiation of *Salmonella* spp.

Chromogenic Substrate Broth**Composition per liter:**

NaCl	10.0g
HEPES (N-[2-Hydroxyethyl] piperazine-N'-[2-ethane-sulfonic acid]) buffer	6.9g
(NH ₄) ₂ SO ₄	5.0g

<i>o</i> -Nitrophenyl- β -D-galactopyranoside.....	0.5g
Solanium	0.5g
MgSO ₄	0.1g
4-Methylumbelliferyl- β -D-glucuronide.....	0.075g
CaCl ₂	0.05g
Na ₂ SO ₃	0.04g
Amphotericin B	1.0mg
MnSO ₄	0.5mg
ZnSO ₄	0.5mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the detection of coliform bacteria based on their hydrolysis of chromogenic substrates by production of β -D-galactopyranosidase. Bacteria that produce β -D-galactopyranosidase turn the medium yellow.

CIN Agar (*Yersinia* Selective Agar)

(Cefsulodin Irgasan Novobiocin Agar)

Composition per liter:

Mannitol.....	20.0g
Agar	12.0g
Pancreatic digest of gelatin	10.0g
Beef extract.....	5.0g
Peptic digest of animal tissue.....	5.0g
Sodium pyruvate.....	2.0g
Yeast extract.....	2.0g
NaCl.....	1.0g
Sodium deoxycholate.....	0.5g
Neutral Red.....	0.03g
Cefsulodin.....	0.015g
Irgasan (triclosan)	4.0mg
Novobiocin.....	2.5mg
Crystal Violet	1.0mg

pH 7.4 $\pm$ 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except cefsulodin and novobiocin, to distilled/deionized water and bring volume to 1.0L. Heat, mixing continuously, until boiling. Do not autoclave. Cool to 45°–50°C. Aseptically add cefsulodin and novobiocin. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and differentiation of *Yersinia enterocolitica* based on mannitol fermentation. *Yersinia enterocolitica* appears as “bull’s eye” colonies with deep red centers surrounded by a transparent periphery.

Clostridia Medium

Composition per liter:

Sodium L-lactate	10.0g
Sodium acetate.....	8.0g
K ₂ HPO ₄	0.5g
(NH ₄) ₂ ·7H ₂ O.....	0.5g
Sodium thioglycolate	0.5g
Yeast extract.....	0.5g
MgSO ₄ ·7H ₂ O	0.1g
FeSO ₄ ·7H ₂ O.....	0.02g
<i>p</i> -Aminobenzoate.....	100.0 μ g
Biotin	0.1 μ g

pH 6.0–7.0 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.0–7.0. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Clostridium* species that ferment lactate and acetate.

Clostridium acidurici Medium

Composition per liter:

Uric acid.....	2.0g
Yeast extract.....	1.0g
K ₂ HPO ₄	0.91g
KOH.....	0.67g
Sodium thioglycolate	0.5g
MgSO ₄ ·7H ₂ O	0.25g
CaCl ₂ ·2H ₂ O	0.015g
FeSO ₄ ·7H ₂ O.....	6.0mg
NaHCO ₃ solution	25.0mL
Sodium thioglycolate solution	25.0mL

pH 7.0–7.5 at 25°C

NaHCO₃ Solution:

Composition per 25.0mL:

NaHCO ₃	5.0g

Preparation of NaHCO₃ Solution: Add NaHCO₃ to distilled/deionized water and bring volume to 25.0mL. Mix thoroughly. Filter sterilize.

Sodium Thioglycolate Solution:

Composition per 25.0mL:

Sodium thioglycolate	0.5g

Preparation of Sodium Thioglycolate Solution: Add sodium thioglycolate to distilled/deionized water and bring volume to 25.0mL. Mix thoroughly. Autoclave solution separately for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add K₂HPO₄ and KOH to distilled/deionized water and bring volume to 1.0L. Add uric acid. Gently heat until boiling. Add the remaining components, except NaHCO₃ and sodium

thioglycolate. Mix thoroughly. Adjust pH to 7.0–7.5. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Add 0.25mL of sterile NaHCO₃ solution and 0.25mL of sterile sodium thioglycolate solution for each 10.0mL of sterile basal medium.

Use: For the cultivation and maintenance of *Clostridium acidurici*, *Clostridium purinolyticum*, and other bacteria that can utilize uric acid as a carbon source.

***Clostridium aerotolerans* Medium**

Composition per liter:

Agar	15.0g
Xylan.....	5.0g
Yeast extract.....	5.0g
Na ₂ CO ₃	4.0g
NaCl.....	0.45g
(NH ₄) ₂ SO ₄	0.45g
K ₂ HPO ₄	0.225g
KH ₂ PO ₄	0.225g
L-Cysteine·HCl·H ₂ O	0.125g
Na ₂ S·9H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.09g
CaCl ₂	0.045g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Prepare medium anaerobically under 100% CO₂. Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Adjust pH to 7.0. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Clostridium aerotolerans*.

***Clostridium* Alginate Medium**

Composition per liter of seawater:

Agar	15.0g
Sodium alginate	10.0g
K ₂ HPO ₄	2.0g
Peptone.....	1.0g
Yeast extract.....	1.0g
Seawater.....	1.0L

pH 7.0–7.5 at 25°C

Preparation of Medium: Add K₂HPO₄ to 1.0L of seawater. Mix thoroughly. Gently heat while stirring to dissolve. Filter solution twice. Add remaining components. Mix thoroughly. Adjust pH to 7.0–7.5. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Clostridium alginolyticum* and other bacteria that can utilize alginate as a carbon source.

***Clostridium aminobutyricum* Medium**

Composition per liter:

K ₂ HPO ₄	7.05g
Yeast extract.....	3.0g
KH ₂ PO ₄	1.29g
MgCl ₂ ·6H ₂ O	0.2g
CaCl ₂ ·2H ₂ O	0.01g
FeCl ₃ ·6H ₂ O	0.01g
Methylene Blue.....	2.0mg
MnSO ₄ ·H ₂ O	1.0mg
Na ₂ MoO ₄ ·2H ₂ O	1.0mg
γ-Aminobutyrate solution	100.0mL
Na ₂ CO ₃ solution	100.0mL
Na ₂ S·9H ₂ O solution.....	50.0mL

pH 7.4–7.7 at 25°C

γ-Aminobutyrate Solution:

Composition per 100.0mL:

γ-Aminobutyrate	5.0g

Preparation of γ-Aminobutyrate Solution:

Add γ-aminobutyrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Na₂CO₃ Solution:

Composition per 100.0mL:

Na ₂ CO ₃	2.0g

Preparation of Na₂CO₃ Solution: Add Na₂CO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Na₂S·9H₂O Solution:

Composition per 50.0mL:

Na ₂ S·9H ₂ O	0.3g

Preparation of Na₂S·9H₂O Solution: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except γ-aminobutyrate solution, Na₂CO₃ solution, and Na₂S·9H₂O solution, to distilled/deionized water and bring volume to 750.0mL. Autoclave for 15 min at 15 psi pressure–121°C. Cool under 80% N₂ + 10% CO₂ + 10% H₂. Aseptically add the sterile γ-aminobutyrate solution, Na₂CO₃ solution, and Na₂S·9H₂O solution. Adjust pH to 7.4–7.7. Distribute using anaerobic technique into tubes or flasks.

Use: For the cultivation and maintenance of *Clostridium aminobutyricum* and other bacteria which can utilize aminobutyric acid as a carbon source.

Clostridium aminobutyricum* Medium*Composition per liter:**

K ₂ HPO ₄	7.0g
γ-Aminobutyric acid	5.0g
Yeast extract.....	3.0g
Agar	1.5g
KH ₂ PO ₄	1.3g
MgCl ₂ ·6H ₂ O	0.2g
CaCl ₂ ·2H ₂ O.....	0.01g
FeCl ₃ ·6H ₂ O.....	0.01g
Na ₂ MoO ₄ ·2H ₂ O	1.0mg
Resazurin	1.0mg
NaHCO ₃ solution	20.0mL
Na ₂ S·9H ₂ O solution.....	20.0mL

pH 7.2 ± 0.2 at 25°C

NaHCO₃ Solution:**Composition per 20.0mL:**

NaHCO ₃	1.0g

Preparation of NaHCO₃ Solution: Add NaHCO₃ to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Na₂S·9H₂O Solution:**Composition per 20.0mL:**

Na ₂ S·9H ₂ O.....	0.3g

Preparation of Na₂S·9H₂O Solution: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except NaHCO₃ solution and Na₂S·9H₂O solution, to distilled/deionized water and bring volume to 960.0mL. Mix thoroughly. Sparge under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically and anaerobically add 20.0mL of sterile NaHCO₃ solution and 20.0mL of sterile Na₂S·9H₂O solution. Mix thoroughly.

Use: For the growth and maintenance of *Clostridium aminobutyricum* and other *Clostridium* species

Clostridium botulinum* Isolation Agar (CBI Agar)*Composition per 1033.0mL:**

Egg yolk agar base.....	900.0mL
Egg yolk emulsion, 50%.....	100.0mL
Cycloserine solution	25.0mL
Sulfamethoxazole solution.....	4.0mL
Trimethoprim solution	4.0mL

pH 7.4 ± 0.2 at 25°C

Egg Yolk Agar Base:**Composition per 900.0mL:**

Pancreatic digest of casein.....	40.0g

Agar	20.0g
Na ₂ HPO ₄	5.0g
Yeast extract.....	5.0g
Glucose	2.0g
NaCl.....	2.0g
MgSO ₄ ·7H ₂ O solution	0.2mL

Preparation of Egg Yolk Agar Base: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

MgSO₄·7H₂O Solution:**Composition per 100.0mL:**

MgSO ₄ ·7H ₂ O	5.0g

Preparation of MgSO₄·7H₂O Solution: Add MgSO₄·7H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Cycloserine Solution:**Composition per 100.0mL:**

Cycloserine	1.0g

Preparation of Cycloserine Solution: Add cycloserine to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Sulfamethoxazole Solution:**Composition per 100.0mL:**

Sulfamethoxazole	1.9g

Preparation of Sulfamethoxazole Solution:

Add sulfamethoxazole to distilled/deionized water and bring volume to 50.0mL. Add sufficient 10% NaOH to dissolve. Bring volume to 100.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Trimethoprim Solution:**Composition per 100.0mL:**

Trimethoprim.....	0.1g

Preparation of Trimethoprim Solution:

Add trimethoprim to distilled/deionized water and bring volume to 50.0mL. Gently heat to 55°C. Add sufficient 0.05N HCl to dissolve. Bring volume to 100.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg	1
NaCl (0.9% solution).....	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to

50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Aseptically add warmed, sterile egg yolk emulsion, 50%, and sterile cycloserine solution, sterile sulfamethoxazole solution, and sterile trimethoprim solution to cooled, sterile egg yolk agar base. Mix thoroughly. Pour into sterile Petri dishes.

Use: For isolation, cultivation, and differentiation based on lipase activity of *Clostridium botulinum* types A, B, and F. *Clostridium botulinum* types A, B, and F appear as raised colonies surrounded by an opaque zone. Other *Clostridium* species and *Clostridium botulinum* type G appear as pinpoint colonies with no opaque zone.

***Clostridium* CK Medium (DSMZ Medium 869)**

Composition per liter:

Pancreatic digest of casein	3.2g
NaCl	0.9g
Papaic digest of soybean meal	0.6g
K ₂ HPO ₄	0.5g
Glucose	0.5g
Resazurin	0.5mg
Glucose solution	10.0mL
pH 5.5 ± 0.2 at 25°C	

Glucose Solution:

Composition per 10.0mL:

Glucose	2.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 100% N₂. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 5.5. Sparge with 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically and anaerobically add 10.0mL sterile glucose solution. Aseptically and anaerobically distribute into tubes or flasks.

Use: For the cultivation of *Clostridium akagii*, *Clostridium acidisoli*, and *Clostridium uliginosum*.

***Clostridium difficile* Agar**

Composition per liter:

<i>Clostridium difficile</i> agar base	920.0mL
<i>Clostridium difficile</i> selective supplement	10.0mL
Horse blood, defibrinated	70.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

***Clostridium difficile* Agar Base:**

Composition per 920.0mL:

Proteose peptone	40.0g
Agar	15.0g
Fructose	6.0g
Na ₂ HPO ₄	5.0g
NaCl	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g

Preparation of *Clostridium difficile* Agar

Base: Add components to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Gently heat to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

***Clostridium difficile* Selective Supplement:**

Composition per 10.0mL:

D-Cycloserine	500.0mg
Cefoxitin	16.0mg

Preparation of *Clostridium difficile* Selective

Supplement: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add 10.0mL of sterile *Clostridium difficile* selective supplement and 70.0mL of sterile, defibrinated horse blood to 920.0mL of cooled, sterile *Clostridium difficile* agar base. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Clostridium difficile*.

***Clostridium difficile* Agar (Cycloserine Cefoxitin Fructose Agar) (CCFA)**

Composition per liter:

Peptic digest of animal tissue	32.0g
Agar	20.0g
Fructose	6.0g
Na ₂ HPO ₄	5.0g
NaCl	2.0g
KH ₂ PO ₄	1.0g
Cycloserine	0.25g
MgSO ₄	0.1g
Neutral Red	0.03g
Cefoxitin solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Cefoxitin Solution:**Composition per 10.0mL:**

Cefoxitin 16.0mg

Preparation of Cefoxitin Solution: Add cefoxitin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile cefoxitin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Clostridium difficile*.

***Clostridium perfringens* Agar, OPSP (Perfringens Agar, OPSP)**

Composition per liter:

Pancreatic digest of casein 15.0g

Agar 10.0g

Liver extract 7.0g

Papaic digest of soybean meal 5.0g

Yeast extract 5.0g

Tris(hydroxymethyl)aminomethane buffer 1.5g

Ferric ammonium citrate 1.0g

Na₂S₂O₅ 1.0g

Antibiotic inhibitor 10.0mL

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Antibiotic Inhibitor:**Composition per 10.0mL:**

Sodium sulfadiazine 0.1g

Oleandomycin phosphate 0.5mg

Polymyxin B 10,000U

Preparation of Antibiotic Inhibitor: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic inhibitor, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the presumptive identification and enumeration of *Clostridium perfringens* in foods.

***Clostridium* Selective Agar (Clostrisel Agar)**

Composition per liter:

Pancreatic digest of casein 17.0g

Agar 14.0g

Glucose 6.0g

Papaic digest of soybean meal 3.0g

NaCl 2.5g

Sodium thioglycolate 1.8g

Sodium formaldehyde sulfoxylate 1.0g

L-Cystine 0.25g

Na₃ 0.15g

Neomycin sulfate 0.15g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Use: For the selective isolation of pathogenic *Clostridium* species from specimens containing mixed flora.

Coagulase Agar Base

Composition per liter:

Agar 25.0g

Brain heart infusion 10.5g

Pancreatic digest of casein 10.5g

D-Mannitol 10.0g

Brain heart infusion 5.0g

NaCl 3.5g

Papaic digest of soybean meal 3.5g

Bromcresol Purple 0.02g

Rabbit plasma 100.0mL

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components, except rabbit plasma, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat, while stirring, until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Add rabbit plasma to a final concentration of 7–15%. Mix thoroughly. Pour into sterile Petri dishes in 18.0mL volume per plate.

Use: For the cultivation and differentiation of *Staphylococcus aureus* from other *Staphylococcus* species based on coagulase production.

Coagulase Mannitol Agar

Composition per liter:

Agar	14.5g
Pancreatic digest of casein	10.5g
D-Mannitol	10.0g
Brain heart infusion	5.0g
NaCl	3.5g
Papaic digest of soybean meal	3.5g
Bromcresol Purple	0.02g
Rabbit plasma with 0.15% EDTA	100.0mL
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except rabbit plasma, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Add rabbit plasma with 0.15% EDTA to a final concentration of 7–15%. Mix thoroughly. Pour into sterile Petri dishes in 18.0mL volume per plate.

Use: For the cultivation and differentiation of *Staphylococcus aureus* from other *Staphylococcus* species based on coagulase production and mannitol fermentation.

Coletsos Medium

Composition per 1625mL:

Potato starch	10.0g
Gelatin	4.0g
Asparagine	2.25g
KH ₂ PO ₄	1.5g
Na-glutamate	1.0g
Na-pyruvate	1.0g
Mg-citrate	0.375g
Litmus	0.25g
Malachite green	0.25g
MgSO ₄	0.15g
Activated carbon	0.1g
Oligonucleotide mixture	3.0mg
Homogenized egg	625.0mL
Glycerol	7.5mL

Egg Mixture:

Composition per liter:

Whole eggs	18–24

Preparation of Egg Mixture: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Separate egg whites from egg yolks. Combine 8 parts egg white with 2 parts egg yolk. Homogenize by shaking. Filter

through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except egg mixture. Bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 625.0mL of egg mixture. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation of *Mycobacterium tuberculosis*.

Coletsos Selective Medium

Composition per 1625mL:

Potato starch	10.0g
Gelatin	4.0g
Asparagine	2.25g
KH ₂ PO ₄	1.5g
Na-glutamate	1.0g
Na-pyruvate	1.0g
Mg-citrate	0.375g
Litmus	0.25g
Malachite green	0.25g
MgSO ₄	0.15g
Activated carbon	0.1g
Oligonucleotide mixture	3.0mg
Homogenized egg	625.0mL
Glycerol	7.5mL
Nalidixic acid solution	1.0mL
Lincomycin solution	1.0mL
Cycloheximide solution	1.0mL

Nalidixic Acid Solution:

Composition per 100.0mL:

Nalidixic acid	0.5g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 100.0mL:

Cycloheximide	1.5g
Ethanol	40.0mL

Preparation of Cycloheximide Solution: Add cycloheximide to 40.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Lincomycin Solution:

Composition per 100.0mL:

Lincomycin	0.5g

Preparation of Lincomycin Solution: Add lincomycin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Egg Mixture Solution:

Composition per liter:

Whole eggs 18–24

Preparation of Egg Mixture Solution: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Separate egg whites from egg yolks. Combine 8 parts egg white with 2 parts egg yolk. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except egg mixture, lincomycin solution, cycloheximide solution, and nalidixic acid solution. Mix thoroughly. Bring volume to 1.0L. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 625.0mL of egg mixture. Mix thoroughly. Aseptically add 1.0mL cycloheximide solution, 1.0mL lincomycin solution, and 1.0mL nalidixic acid solution. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the isolation and cultivation of *Mycobacterium tuberculosis*.

Coli ID

Composition per liter:

Proprietary

Source: This medium is available from bioMérieux.

Use: A selective chromogenic medium for the detection and enumeration of *E. coli* at 44°C, and simultaneous enumeration of *E. coli* and other coliforms at 37°C, from food products.

Coliform Agar, Chromocult® (Chromocult Coliform Agar)

Composition per liter:

Agar 10.0g
NaCl 5.0g
Peptone 3.0g
Na₂HPO₄ 2.7g
NaH₂PO₄ 2.2g
Tryptophan 1.0g
Na-pyruvate 1.0g

Chromogenic mixture 0.4g
Tergitol 7 0.15g
pH 7.0 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix well and warm gently until dissolved. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes. Some turbidity may occur, but this does not effect the performance

Use: For the detection of *E. coli* and coliform bacteria in foods. The interaction of selected peptones, pyruvate, sorbitol, and phosphate buffer guarantees rapid colony growth, even for sublethally injured coliforms. The growth of Gram-positive bacteria as well as some Gram-negative bacteria is largely inhibited by the content of Tergitol 7 which has no negative effect on the growth of the coliform bacteria. A combination of two chromogenic substrates which allow for the simultaneous detection of total coliforms and *E. coli*. The characteristic enzyme for coliforms, β-D-cleaves the Salmon-GAL substrate and causes a salmon to red color of the coliform colonies. The substrate X-glucuronide is used for the identification of β-D-glucuronidase, which is characteristic for *E. coli*. *E. coli* cleaves both Salmon-GAL and X-glucuronide, so that positive colonies take on a dark-blue to violet color. These are easily distinguished from other coliform colonies which have a salmon to red color. As part of an additional confirmation of *E. coli*, the inclusion of tryptophan improves the indole reaction, thereby increasing detection reliability when it is used in combination with the Salmon-GAL and X-glucuronide reaction.

Coliform Agar ES, Chromocult (Chromocult Coliform Agar ES) (Chromocult Enhanced Selectivity Agar)

Composition per liter:

Agar 10.0g
MOPS 10.0g
KCl 7.5g
Peptone 5.0g
Bile salts 1.15g
Na-propionate 0.5g
6-Chloro-3-indoxyl-β-D-galactopyranoside 0.15g
5-Bromo-4-chloro-3-indoxyl-
β-D-glucuronic acid 0.1g
Isopropyl-β-D-thiogalactopyranoside 0.1g
pH 7.0 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until components are completely dissolved (approximately 45 min). Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. The plates should be clear and colorless.

Use: For the detection of *E. coli* and total coliforms. The combination of suitable peptones and the buffering using MOPS allow rapid growth of coliforms and an optimal transformation of the chromogenic substrates. The amount of bile salts and propionate largely inhibit growth of Gram-positive and Gram-negative accompanying flora. The simultaneous detection of total coliforms and *E. coli* is achieved using the combination of two chromogenic substrates. The substrate Salmon™-β-D-GAL is split by β-D-galactosidase, characteristic for coliforms, resulting in a salmon to red coloration of coliform colonies. The detection of the β-D-glucuronidase, characteristic for *E. coli*, is cleaved via the substrate X-β-D-glucuronide, causing a blue coloration of positive colonies. As *E. coli* splits Salmon-β-D-GAL as well as X-β-D-glucuronide, the colonies turn to a dark violet color and can be easily differentiated from the other coliforms being salmon-red.

Coliform Medium (CM)

Composition per liter:

Bile salts No. 3	20.0g
Lactose	20.0g
Proteose peptone No. 3	10.0g
Yeast extract	6.0g
Sodium lauryl sulfate	1.0g
Sodium deoxycholate	0.1g
Bromcresol Purple solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.35g
NaOH (0.1N solution)	2.0mL

Preparation of Bromcresol Purple Solution: Combine Bromcresol Purple and NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Preparation of Medium: Add components, except Bromcresol Purple solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Add 10.0mL of Bromcresol Purple solution. Mix thoroughly. Adjust pH to 7.0 with 1N HCl. Distribute into flasks in 95.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of coliform microorganisms from cream.

Coliform Medium (CM)

Composition per liter:

Bile salts No. 3	20.0g
Lactose	20.0g
Proteose peptone No. 3	10.0g
Yeast extract	6.0g
Sodium lauryl sulfate	1.0g
Sodium deoxycholate	0.1g
Bromcresol Purple solution	10.0mL
pH 6.8 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.35g
NaOH (0.1N solution)	2.0mL

Preparation of Bromcresol Purple Solution:

Combine Bromcresol Purple and NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Preparation of Medium: Add components, except Bromcresol Purple solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Add 10.0mL of Bromcresol Purple solution. Mix thoroughly. Adjust pH to 6.8 with 1N NH₄OH. Distribute into flasks in 95.0mL volumes. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of coliform microorganisms from yogurt and raw milk.

Coliform Medium, Modified (MCM)

Composition per liter:

Lactose	20.0g
Tris(hydroxymethyl)aminomethane buffer	12.1g
Proteose peptone No. 3	10.0g
Yeast extract	6.0g
Bile salts No. 3	1.0g
Sodium lauryl sulfate	1.0g
Sodium deoxycholate	0.1g
Bromcresol Purple solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.35g
NaOH (0.1N solution)	2.0mL

Preparation of Bromcresol Purple Solution:

Combine Bromcresol Purple and NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Preparation of Medium: Add components, except Bromcresol Purple solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Add 10.0mL of Bromcresol Purple solution. Mix thoroughly. Adjust pH to 7.0 with 1N HCl. Distribute into flasks in 95.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of coliform microorganisms from cream.

Coliform Medium, Modified (MCM)

Composition per liter:

Lactose	20.0g
Tris(hydroxymethyl)aminomethane buffer	12.1g
Proteose peptone No. 3	10.0g
Yeast extract	6.0g
Bile salts No. 3	1.0g
Sodium lauryl sulfate	1.0g
Sodium deoxycholate	0.1g
Bromcresol Purple solution	10.0mL
pH 6.8 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.35g
NaOH (0.1N solution)	2.0mL

Preparation of Bromcresol Purple Solution:

Combine Bromcresol Purple and NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Preparation of Medium: Add components, except Bromcresol Purple solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Add 10.0mL of Bromcresol Purple solution. Mix thoroughly. Adjust pH to 6.8 with 1N NH₄OH. Distribute into flasks in 95.0mL volumes. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of coliform microorganisms from yogurt.

Colonization Medium

Composition per 700.0mL:

Mannose	1.0g
Pancreatic digest of gelatin	0.2g
Brain heart, solids from infusion	0.08g
Peptic digest of animal tissue	0.08g
NaCl	0.07g
Glucose	0.04g
Na ₂ HPO ₄	0.03g
Bile salts No. 3	0.1g
Dulbecco's phosphate-buffered saline	700.0mL
pH 7.4 ± 0.2 at 25°C	

Dulbecco's Phosphate-Buffered Saline:

Composition per liter:

NaCl	8.0g
Na ₂ HPO ₄ ·7H ₂ O	2.16g
KCl	0.2g
KH ₂ PO ₄	0.2g
CaCl ₂	0.1g
MnCl ₂ ·6H ₂ O	0.1g

Preparation of Dulbecco's Phosphate-Buffered Saline: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Filter sterilize. Aseptically distribute into sterile tubes or flasks.

Use: For the differentiation of enterotoxigenic *Escherichia coli* from foods based on the HeLa cell test for colonization.

Congo Red Agar (CR Agar)

Composition per liter:

GC agar base	890.0mL
Hemoglobin solution	100.0mL
Supplement solution	10.0mL
Congo Red (0.01% solution)	0.1mL
pH 7.2 ± 0.2 at 25°C	

GC Agar Base:

Composition per 890.0mL:

Agar	10.0g
Pancreatic digest of casein	7.5g
Peptic digest of animal tissue	7.5g
NaCl	5.0g
K ₂ HPO ₄	4.0g
Cornstarch	1.0g
KH ₂ PO ₄	1.0g

Preparation of GC Agar Base: Add components to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Hemoglobin Solution:

Composition per 100.0mL:

Hemoglobin	2.0g

Preparation of Hemoglobin Solution: Add hemoglobin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Congo Red Solution:

Composition per 100.0mL:

Congo Red	0.01g

Preparation of Congo Red Solution: Add Congo Red to 100.0mL of distilled/deionized water. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Supplement Solution:

Composition per liter:

Glucose	100.0g
L-Cysteine-HCl.....	25.9g
L-Glutamine.....	10.0g
L-Cystine	1.1g
Adenine.....	1.0g
Nicotinamide adenine dinucleotide	0.25g
Vitamin B ₁₂	0.1g
Thiamine pyrophosphate.....	0.1g
Guanine-HCl	0.03g
Fe(NO ₃) ₃ ·6H ₂ O	0.02g
p-Aminobenzoic acid.....	0.013g
Thiamine-HCl	3.0mg

Preparation of Supplement Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Source: The supplement solution IsoVitaleX® enrichment is available from BD Diagnostic Systems. This enrichment may be replaced by supplement VX from BD Diagnostic Systems.

Preparation of Medium: To 890.0mL of sterile, cooled GC agar base, aseptically add 100.0mL of sterile, cooled hemoglobin solution, 10.0mL of sterile supplement solution, and 0.1mL of sterile Congo Red solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the isolation and differentiation of virulent and avirulent strains of *Shigella*, *Vibrio cholerae*, and *Escherichia coli*.

Congo Red Agar (CR Agar)

Composition per liter:

Soybean-casein digest agar.....	890.0mL
Hemoglobin solution.....	100.0mL
Supplement solution	10.0mL
Congo Red (0.01% solution)	0.1mL
pH 7.3 ± 0.2 at 25°C	

Soybean-Casein Digest Agar:

Composition per 890.0mL:

Pancreatic digest of casein.....	17.0g
Agar	15.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g

Preparation of Soybean-Casein Digest Agar:

Add components to distilled/deionized water and

bring volume to 890.0mL. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Hemoglobin Solution:

Composition per 100.0mL:

Hemoglobin	2.0g

Preparation of Hemoglobin Solution: Add hemoglobin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Congo Red Solution:

Composition per 100.0mL:

Congo Red	0.01g

Preparation of Congo Red Solution: Add Congo Red to 100.0mL of distilled/deionized water. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Supplement Solution:

Composition per liter:

Glucose	100.0g
L-Cysteine-HCl.....	25.9g
L-Glutamine	10.0g
L-Cystine	1.1g
Adenine.....	1.0g
Nicotinamide adenine dinucleotide	0.25g
Vitamin B ₁₂	0.1g
Thiamine pyrophosphate	0.1g
Guanine-HCl.....	0.03g
Fe(NO ₃) ₃ ·6H ₂ O.....	0.02g
p-Aminobenzoic acid.....	0.013g
Thiamine-HCl	3.0mg

Preparation of Supplement Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 890.0mL of sterile, cooled soybean-casein digest agar, aseptically add 100.0mL of sterile, cooled hemoglobin solution, 10.0mL of sterile supplement solution, and 0.1mL of sterile Congo Red solution. Mix thoroughly. Pour into sterile Petri dishes.

Source: The supplement solution IsoVitaleX enrichment is available from BD Diagnostic Systems. This enrichment may be replaced by supplement VX from BD Diagnostic Systems.

Use: For the isolation and differentiation of virulent and avirulent strains of *Shigella*, *Vibrio cholerae*, and *Escherichia coli*. Used for the detection and differentiation of “iron-responsive” avirulent mutants.

Congo Red BHI Agarose Medium**Composition per liter:**

Agarose	15.0g
Pancreatic digest of gelatin	14.5g
Brain heart, solids from infusion	6.0g
Peptic digest of animal tissue.....	6.0g
NaCl.....	5.0g
Glucose	3.0g
Na ₂ HPO ₄	2.5g
Congo Red	0.075g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the isolation, cultivation, and detection of virulent strains of *Yersinia enterocolitica*.

**Congo Red BHI Agarose Medium
(CRBHO Medium)
(BAM M41)**
Composition per liter:

Agarose	12.0g
Pancreatic digest of gelatin	14.5g
Brain heart, solids from infusion	6.0g
Peptic digest of animal tissue.....	6.0g
NaCl.....	5.0g
Glucose	3.0g
Na ₂ HPO ₄	2.5g
MgCl ₂	1.0g
Congo Red solution	20.0mL

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes.

Congo Red Solution:**Composition per 100.0mL:**

Congo Red	375.0mg

Preparation of Congo Red Solution: Add Congo Red to 100.0mL of distilled/deionized water. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Use: For the isolation, cultivation, and detection of virulent strains of *Yersinia enterocolitica*.

**Congo Red Magnesium
Oxalate Agar
(CRMOX Agar)**
Composition per liter:

Solution 1	825.0mL
Solution 2.....	80.0mL
Solution 3.....	80.0mL
Solution 4.....	10.0mL
Solution 5.....	5.0mL

pH 7.3 ± 0.2 at 25°C

Solution 1:**Composition per 825.0mL:**

Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g
NaCl.....	5.0g

pH 7.3 ± 0.2 at 25°C

Preparation of Solution 1: Add components to distilled/deionized water and bring volume to 825.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat.

Solution 2:**Composition per liter:**

MgCl ₂ ·6H ₂ O	50.8g

Preparation of Solution 2: Add MgCl₂·6H₂O to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution 3:**Composition per liter:**

Sodium oxalate	33.2g

Preparation of Solution 3: Add sodium oxalate to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution 4:**Composition per 100.0mL:**

D-Galactose	20.0g

Preparation of Solution 4: Add D-galactose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Solution 5:**Composition per 10.0mL:**

Congo Red	0.1g

Preparation of Solution 5: Add Congo Red to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Aseptically combine 80.0mL of sterile solution 2, 80.0mL of sterile solution 3, 10.0mL of sterile solution 4, and 5.0mL of sterile solution 5. Mix thoroughly. Warm to 50°C. Add this mixture to 825.0mL of cooled, sterile solution 1. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation and identification of pathogenic serotypes of *Yersinia enterocolitica*. For the determination of whether *Yersinia* strains contain the *Yersinia* virulence plasmid.

Conradi Drigalski Agar

Composition per liter:

Agar	15.0g
Casein.....	10.0g
Lactose	10.0g
Peptone.....	10.0g
NaCl.....	5.0g
Bromcresol Purple	0.03g
Crystal Violet	4.0mg

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of Gram-negative enteric bacilli.

Cooke Rose Bengal Agar

Composition per liter:

Agar	20.0g
Glucose	10.0g
Enzymatic hydrolysate of soybean meal	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Rose Bengal	35.0mg

pH 6.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation of fungi.

Cooked Meat Medium (BAM M42)

Composition per liter:

Beef heart.....	454.0g
Proteose peptone.....	20.0g
NaCl.....	5.0g
Glucose	2.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Finely chop beef heart. Add approximately 1.5g of heart particles to test tubes. Add remaining components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Slowly cool tubes to prevent expulsion of meat particles. Steam the sterilized medium and cool, without agitation, just before use.

Use: For the cultivation and maintenance of anaerobic microorganisms.

Cooked Meat Medium

Composition per liter:

Heart muscle	454.0g
Beef extract.....	10.0g
Peptone	10.0g
NaCl.....	5.0g
Glucose	2.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Finely chop beef heart. Add approximately 1.5g of heart particles to test tubes. Add remaining components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Slowly cool tubes to prevent expulsion of meat particles.

Use: For the cultivation and maintenance of aerobic and anaerobic microorganisms. For the cultivation of anaerobes, especially pathogenic *Clostridium* spp.

Cooked Meat Medium

Composition per liter:

Heart tissue granules.....	98.0g
Peptic digest of animal tissue	20.0g
NaCl.....	5.0g
Glucose	2.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add approximately 1.0g of heart tissue granules to test tubes. Add remaining components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Slowly cool tubes to prevent expulsion of meat particles.

Use: For the cultivation of anaerobes, especially pathogenic clostridia.

Cooked Meat Medium, Modified

Composition per liter:

Cooked meat medium	66.0g
Solution A	1.0L
pH 6.8 ± 0.2 at 25°C	

Solution A:

Composition per liter:

Pancreatic digest of casein	10.0g
Glucose	2.0g
Soluble starch	1.0g
Sodium thioglycolate	1.0g
Neutral Red (1% aqueous)	5.0mL

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved.

Preparation of Medium: Add 1.0g of cooked meat medium to each of 66 test tubes. Add 15.0mL of solution A to each test tube. Allow meat particles to rehydrate. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of anaerobic microorganisms.

Cooked Meat Medium, Modified (BAM M43)

Composition per tube:

Cooked meat medium	1.0g
Diluent	1.0L
pH 6.8 ± 0.2 at 25°C	

Cooked Meat Medium:

Composition per 481g:

Beef heart	454.0g
Proteose peptone	20.0g
NaCl	5.0g
Glucose	2.0g

Source: Cooked meat medium is available in dehydrated form from BD Diagnostic Systems.

Diluent:

Composition per liter:

Pancreatic digest of casein	10.0g
Glucose	2.0g
Soluble starch	1.0g

Sodium thioglycolate	1.0g
Neutral Red (1% aqueous)	5.0mL

Preparation of Diluent: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved.

Preparation of Medium: Add 1.0g of dehydrated cooked meat medium and 15.0mL diluent to 20 × 150mm test tubes. Let meat particles rehydrate. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C.

Cornmeal Agar (ATCC Medium 307)

Composition per liter:

Cornmeal	50.0g
Agar	7.5g

Preparation of Medium: Add cornmeal to distilled/deionized water and bring volume to 800.0mL. Leave overnight in refrigerator. Heat to 60°C for 1 hr. Bring volume to 1.0L with distilled/deionized water. Add agar. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of numerous fungi.

Cornmeal Agar (CMA)

Composition per liter:

Agar	20.0g
Cornmeal polenta	15.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add Cornmeal polenta to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through Whatman #1 filter paper. Add agar to filtrate. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of many filamentous fungi.

Cornmeal Agar

Composition per liter:

Agar	15.0g
Cornmeal, solids from infusion	2.0g
pH 5.6–6.0 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fungi.

Cornmeal Agar with Dextrose

Composition per liter:

Agar	15.0g
Cornmeal, solids from infusion	2.0g
Glucose	2.0g
Tween 80	1.0g
pH 5.6–6.0 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of various fungi.

Cornmeal and V8 Juice Agar (ATCC Medium 309)

Composition per liter:

Agar	7.5g
CaCO ₃	3.0g
Cornmeal Extract	800.0mL
V8 Juice	200.0mL
pH 5.6–6.0 at 25°C	

Cornmeal Extract:

Composition per 800.0mL:

Yellow cornmeal	50.0g

Preparation of Cornmeal Infusion: Add 50.0g of yellow Cornmeal to 800.0mL of water. Leave in refrigerator overnight. Heat Cornmeal mixture to 60°C for 1 hr. Filter out Cornmeal through cheese cloth. Bring volume back to 800.0mL.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of fungi.

Cornmeal Agar, Quarter-Strength (ATCC Medium 2221)

Composition per liter:

Agar	15.0g
Cornmeal Infusion	250.0mL
pH 5.6–6.0 at 25°C	

Cornmeal Infusion:

Composition per liter:

Yellow cornmeal	50.0g

Preparation of Cornmeal Infusion: Add cornmeal to distilled/deionized water and bring volume to 1.0L. Gently heat and bring to boiling. Simmer for 10 min. Filter through cheesecloth. Return volume to 1.0 liter.

Preparation of Medium: Add agar to 250.0mL cornmeal infusion and bring volume to 1.0L with distilled/deionized water. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of fungi.

Creatinine/NMH Medium

Composition per 1100.0mL:

Yeast extract	5.0g
NaCl	1.0g
NaHCO ₃	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Na ₂ S·9H ₂ O	0.5g
MnCl ₂ ·4H ₂ O	0.06g
CaSO ₄ ·2H ₂ O	0.05g
FeSO ₄ ·7H ₂ O	0.01g
Na ₂ SeO ₃ ·5H ₂ O	26.0μg
Vitamin B ₁₂	20.0μg
Resazurin	1.0mg
Phosphate solution	100.0mL
Creatinine solution	100.0mL
Trace elements solution SL-4	10.0mL
Vitamin solution	10.0mL
L-Cysteine-HCl solution	10.0mL
Na ₂ S·9H ₂ O solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Phosphate Solution:

Composition per 100.0mL:

K ₂ HPO ₄	5.33g
KH ₂ PO ₄	2.64g

Preparation of Phosphate Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Sparge with 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C.

Creatinine Solution:

Composition per 100.0mL:

Creatinine	5.5g

Preparation of Creatinine Solution: Add creatinine to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Sparge with 100% N₂. Filter sterilize.

Trace Elements Solution SL-4:**Composition** per liter:

EDTA	0.5g
FeSO ₄ ·7H ₂ O	0.2g
Trace elements solution SL-6	100.0mL

Trace Elements Solution SL-6:**Composition** per liter:

MnCl ₂ ·4H ₂ O	0.5g
H ₃ BO ₃	0.3g
CoCl ₂ ·6H ₂ O	0.2g
ZnSO ₄ ·7H ₂ O	0.1g
Na ₂ MoO ₄ ·2H ₂ O	0.03g
NiCl ₂ ·6H ₂ O	0.02g
CuCl ₂ ·2H ₂ O	0.01g

Preparation of Trace Elements Solution SL-6:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Trace Elements Solution SL-4:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Vitamin Solution:**Composition** per liter:

Pyridoxine-HCl	10.0mg
Calcium DL-pantothenate	5.0mg
Lipoic acid	5.0mg
Nicotinic acid	5.0mg
<i>p</i> -Aminobenzoic acid	5.0mg
Riboflavin	5.0mg
Thiamine-HCl	5.0mg
Biotin	2.0mg
Folic acid	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

L-Cysteine-HCl Solution:**Composition** per 10.0mL:

L-Cysteine-HCl	0.5g

Preparation of L-Cysteine-HCl Solution:

Add L-cysteine-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C.

Na₂S·9H₂O Solution:**Composition** per 10.0mL:

Na ₂ S·9H ₂ O	0.5g

Preparation of Na₂S·9H₂O Solution: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except creatinine solution, phosphate solution, L-cysteine-HCl solution, and Na₂S·9H₂O solution, to distilled/deionized water and bring volume to 880.0mL. Mix thoroughly. Adjust pH to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically and anaerobically add 100.0mL of sterile phosphate solution, 10.0mL of sterile L-cysteine-HCl solution, and 10.0mL of sterile Na₂S·9H₂O solution. Immediately prior to use, aseptically and anaerobically add 100.0mL of sterile creatinine solution. Mix thoroughly. Aseptically and anaerobically distribute into tubes or bottles.

Use: For the cultivation and maintenance of *Clostridium* species.

CreDm1 Medium**Composition** per 1002.0mL:

Solution A	980.0mL
Solution D	10.0mL
Solution E	10.0mL
Solution B	1.0mL
Solution C	1.0mL

pH 6.7–6.9 at 25°C

Solution A:**Composition** per 980.0mL:

KH ₂ PO ₄	1.4g
NH ₄ Cl	0.5g
MgCl ₂ ·6H ₂ O	0.2g
CaCl ₂ ·2H ₂ O	0.15g
Yeast extract	50.0mg

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution B (Trace Elements Solution SL-10):**Composition** per liter:

FeCl ₂ ·4H ₂ O	1.5g
CoCl ₂ ·6H ₂ O	190.0mg
MnCl ₂ ·4H ₂ O	100.0mg
ZnCl ₂	70.0mg
Na ₂ MoO ₄ ·2H ₂ O	36.0mg
NiCl ₂ ·6H ₂ O	24.0mg
H ₃ BO ₃	6.0mg
CuCl ₂ ·2H ₂ O	2.0mg
HCl (25% solution)	10.0mL

Preparation of Solution B (Trace Elements Solution SL-10): Add FeCl₂·4H₂O to 10.0mL of HCl solution. Mix thoroughly. Add distilled/deionized water and bring volume to 1.0L. Add remaining components. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution C (Selenite-Tungstate Solution):**Composition** per liter:

NaOH	0.5g
Na ₂ WO ₄ ·2H ₂ O	4.0mg
Na ₂ SeO ₃ ·5H ₂ O	3.0mg

Preparation of Solution C (Selenite-Tungstate Solution): Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution D (Vitamin Solution):**Composition** per liter:

Pyridoxine-HCl	10.0mg
Calcium DL-pantothenate	5.0mg
Lipoic acid	5.0mg
Nicotinic acid	5.0mg
p-Aminobenzoic acid	5.0mg
Riboflavin	5.0mg
Thiamine-HCl	5.0mg
Biotin	2.0mg
Folic acid	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Solution D (Vitamin Solution): Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Solution E:**Composition** per 10.0mL:

Disodium-DL-malate	1.6g

Preparation of Solution E: Add disodium-DL-malate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Aseptically combine 980.0mL of sterile solution A with 1.0mL of sterile solution B, 1.0mL of sterile solution C, 10.0mL of sterile solution D, and 10.0mL of sterile solution E, in that order. Mix thoroughly. Adjust pH to 6.7–6.9. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Campylobacter* species.

Crossley Milk Medium**Composition** per liter:

Skim milk powder	100.0g
Peptone	10.0g
Bromocresol Purple	0.1g
pH 5.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to a very small volume of distilled/deionized water and

mix to a paste. Gradually add more distilled/deionized water and bring volume to 1.0L. Distribute in 10.0mL volumes into tubes. Autoclave for 5 min at 15 psi pressure–121°C.

Use: For the routine examination of canned food samples for anaerobic bacteria.

Crystal Violet Agar**Composition** per liter:

Agar	15.0g
Lactose	10.0g
Proteose peptone	5.0g
Beef extract	3.0g
Crystal Violet	3.3mg
pH 6.8 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of pathogenic from non-pathogenic staphylococci. Hemolytic and coagulating strains of *Staphylococcus aureus* appear as purple or yellow colonies. Nonhemolytic and noncoagulating strains of *Staphylococcus* species appear as white colonies.

**Crystal Violet Pectate Medium
(CVP Medium)****Composition** per liter:

Sodium polypectate	9.0g
Agar	2.0g
NaNO ₃	1.0g
NaOH (1N solution)	4.5mL
CaCl ₂ ·H ₂ O (10% solution)	3.0mL
Crystal Violet (0.075% solution)	1.0mL
Sodium lauryl sulfate (10% solution)	0.5mL
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: In a preheated blender, add 500.0mL of boiling distilled/deionized water and the components, except sodium polypectate and sodium lauryl sulfate solution. Blend at high speed for 15 sec. Continue blending at low speed and slowly add 9.0g of sodium polypectate. Pour the incomplete medium into a 2L flask and add 0.5mL of sodium lauryl sulfate solution. Mix thoroughly. Cap flask with an aluminum foil seal rather than cotton. Autoclave for 25 min at 15 psi pressure–121°C. Pour medium quickly into sterile Petri dishes. Allow plates to dry at 25°C for 48 hr before use.

Use: For the cultivation of pectinolytic microorganisms, such as *Erwinia* species, from foods.

CS Vitamin B₁₂ Agar**Composition per liter:**

Glucose	20.0g
K ₂ SO ₄	20.0g
Agar	15.0g
Sodium acetate	12.0g
Vitamin assay casamino acids	10.0g
Papaic digest of soybean meal	5.0g
Sodium thioglycolate	1.7g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
Ribonucleic acid	1.0g
Sorbitan monooleate complex	1.0g
MgSO ₄ ·7H ₂ O	0.4g
DL-Tryptophan	0.2g
L-Cystine	0.2g
FeSO ₄	0.02g
MgSO ₄ ·7H ₂ O	0.02g
NaCl	0.02g
Adenine sulfate	0.018g
Guanine-HCl	0.012g
Uracil	0.01g
Xanthine	0.01g
Pyridoxal	4.0mg
Pyridoxine	4.0mg
Calcium pantothenate	2.0mg
Niacin	2.0mg
Riboflavin	2.0mg
Thiamine-HCl	2.0mg
Folic acid	1.0mg
Biotin	1.0μg
<i>Lactobacillus leichmannii</i> suspension	1.0mL
pH 6.2 ± 0.1 at 25°C	

Preparation of Medium: Add components, except *Lactobacillus leichmannii* suspension, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Inoculate medium with 1.0mL of *Lactobacillus leichmannii* suspension. Mix thoroughly. Pour into sterile 150mm Petri dishes in 50.0mL volumes. Allow agar surface to dry before using.

Use: For the microbiological assay of vitamin B₁₂ by the cup plate or disk method using *Lactobacillus leichmannii* as the test microorganism.

**CYA Agar with Arginine
and *p*-Aminobenzoic Acid
(ATCC Medium 2033)**

Composition per liter:

Sucrose	30.0g
Agar	15.0g
Yeast extract	5.0g
NaNO ₃	3.0g

K ₂ HPO ₄	1.0g
KCl	0.5g
MgSO ₄ ·7H ₂ O	0.5g
Arginine	0.2g
FeSO ₄ ·7H ₂ O	0.01g
<i>p</i> -Aminobenzoic acid	1.0mg
pH 7.3 ± 0.2 at 25°C	

Sucrose Solution:**Composition per 100.0mL:**

Sucrose	30.0g

Preparation of Sucrose Solution: Add sucrose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Add components, except sucrose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add sterile sucrose solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of heat-resistant filamentous fungi (molds) from foods.

Czapek Dox Agar, Modified**Composition per liter:**

Sucrose	30.0g
Agar	12.0g
NaNO ₃	2.0g
Magnesium glycerophosphate	0.5g
KCl	0.5g
K ₂ SO ₄	0.35g
FeSO ₄	0.01g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of numerous fungal species.

Czapek Dox Broth**Composition per liter:**

Sucrose	30.0g
NaNO ₃	3.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
KCl	0.5g
FeSO ₄ ·7H ₂ O	0.01g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a variety of fungal and bacterial species that can use nitrate as sole nitrogen source.

Czapek Dox Liquid Medium, Modified

Composition per liter:

Sucrose.....	30.0g
NaNO ₃	2.0g
Magnesium glycerophosphate	0.5g
KCl.....	0.5g
K ₂ SO ₄	0.35g
FeSO ₄	0.01g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fungi and bacteria capable of utilizing sodium nitrate as the sole source of nitrogen.

Czapek Solution Agar

Composition per liter:

Sucrose.....	30.0g
Agar	15.0g
NaNO ₃	2.0g
K ₂ HPO ₄	1.0g
KCl.....	0.5g
MgSO ₄ ·7H ₂ O	0.5g
FeSO ₄ ·7H ₂ O.....	0.01g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Aspergillus*, *Penicillium*, and other fungi. For the cultivation and maintenance of microorganisms that can utilize nitrate as sole nitrogen source.

Czapek Solution Agar with Sucrose

Composition per liter:

Sucrose.....	200.0g
Agar	20.0g
NaNO ₃	3.0g
K ₂ HPO ₄	1.0g
KCl.....	0.5g
MgSO ₄ ·7H ₂ O	0.5g
FeSO ₄ ·7H ₂ O.....	10.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of osmophilic fungi.

Czapek Yeast Autolysate Agar (CYA Agar)/(Czapek Agar)

Composition per liter:

Sucrose.....	30.0g
Agar	15.0g
Yeast extract.....	5.0g
NaNO ₃	3.0g
K ₂ HPO ₄	1.0g
KCl.....	0.5g
MgSO ₄ ·7H ₂ O	0.5g
FeSO ₄ ·7H ₂ O.....	0.01g
pH 7.3 ± 0.2 at 25°C	

Sucrose Solution:

Composition per 100.0mL:

Sucrose.....	30.0g

Preparation of Sucrose Solution: Add sucrose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Add components, except sucrose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add sterile sucrose solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of heat-resistant filamentous fungi (molds) from foods.

Czapek Yeast Extract Agar

Composition per liter:

Sucrose.....	30.0g
Agar	15.0g
Yeast extract.....	5.0g

K ₂ HPO ₄	1.0g
Czapek concentrate	10.0mL

Czapek Concentrate:**Composition per liter:**

NaNO ₃	30.0g
KCl.....	5.0g
MgSO ₄ ·7H ₂ O	5.0g
FeSO ₄ ·7H ₂ O	0.1g
ZnSO ₄ ·7H ₂ O	0.1g
CuSO ₄ ·5H ₂ O	0.05g

Preparation of Czapek Concentrate: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Aspergillus niger*, which is used for the production of food enzymes and citric acid and a common food contaminant.

Czapek Yeast Extract Agar**Composition per liter:**

Sucrose.....	30.0g
Agar	15.0g
Yeast extract.....	5.0g
NaNO ₃	3.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
KCl.....	0.5g
FeSO ₄ ·7H ₂ O	0.01g
Trace metal solution.....	1.0mL

pH 7.3 ± 0.2 at 25°C

Trace Metal Solution:**Composition per 100.0mL:**

ZnSO ₄ ·7H ₂ O	1.0g
CuSO ₄ ·5H ₂ O	0.5g

Preparation of Trace Metal Solution: Add components to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of fungal and bacterial species that can use nitrate as sole nitrogen source.

DCLS Agar (Deoxycholate Citrate Lactose Sucrose Agar)

Composition per liter:

Agar	12.0g
Sodium citrate·3H ₂ O.....	10.5g
Lactose.....	5.0g
Na ₂ S ₂ O ₃	5.0g
Sucrose.....	5.0g
Pancreatic digest of casein.....	3.5g
Peptic digest of animal tissue	3.5g
Beef extract.....	3.0g
Sodium deoxycholate.....	2.5g
Neutral Red.....	0.03g

pH 7.2 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not overheat. Do not autoclave. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the selective isolation of *Salmonella* species, *Shigella* species, and *Vibrio* species.

Decarboxylase Basal Medium (BAM M44)

Composition per liter:

Peptone or gelysate	5.0g
Yeast extract.....	3.0g
Glucose	1.0g
Bromocresol Purple	0.02g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure–121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate the amino acid. As the basal medium for arginine broth, lysine broth, and ornithine broth. Bacteria that decarboxylate arginine, lysine, or ornithine turn the medium turbid purple. The unsupplemented decarboxylase basal medium is used as a control.

Decarboxylase Basal Medium with NaCl (BAM M44)

Composition per liter:

Peptone or gelysate	5.0g
Yeast extract	3.0g
Glucose	1.0g
Bromcresol Purple	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure–121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of *Vibrio* spp. based on their ability to decarboxylate the amino acid. As the basal medium for arginine broth, lysine broth, and ornithine broth. Bacteria that decarboxylate arginine, lysine, or ornithine turn the medium turbid purple. The unsupplemented decarboxylase basal medium is used as a control.

Decarboxylase Base, Møller

Composition per liter:

Amino acid	10.0g
Beef extract	5.0g
Peptone	5.0g
Glucose	0.5g
Bromcresol Purple	0.01g
Cresol Red	5.0mg
Pyridoxal	5.0mg
Mineral oil	200.0mL
pH 6.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except mineral oil, to distilled/deionized water and bring volume to 1.0L. For amino acid, use L-arginine, L-lysine, or L-ornithine. Mix thoroughly. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave medium and mineral oil separately for 15 min at 15 psi pressure–121°C. After inoculation, overlay medium with 1.0mL of sterile mineral oil per tube.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate the amino acid. Bacteria that decarboxylate arginine, lysine, or ornithine turn the medium turbid purple.

Decarboxylase Medium Base, Falkow

Composition per liter:

Amino acid	5.0g
Peptone	5.0g
Yeast extract	3.0g
Glucose	1.0g
Bromcresol Purple	0.02g
Mineral oil	200.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except mineral oil, to distilled/deionized water and bring volume to 1.0L. For amino acid, use L-arginine, L-lysine, or L-ornithine. Mix thoroughly. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave medium and mineral oil separately for 15 min at 15 psi pressure–121°C. After inoculation, overlay medium with 1.0mL of sterile mineral oil per tube.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate the amino acid. Bacteria that decarboxylate arginine, lysine, or ornithine turn the medium turbid purple.

Decarboxylase Medium, Ornithine Modified

Composition per liter:

L-Ornithine	10.0g
Meat peptone	5.0g
Yeast extract	3.0g
Bromcresol Purple solution	5.0mL
pH 5.5 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.2g
Ethanol	50.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 5.5 with HCl or NaOH. Distribute into screw-capped tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate ornithine. Bacteria that decarboxylate ornithine turn the medium turbid purple.

Demi-Fraser Broth**Composition per liter:**

NaCl.....	20.0g
Tryptose	10.0g
Na ₂ HPO ₄	9.6g
Beef extract.....	5.0g
Yeast extract.....	5.0g
LiCl.....	3.0g
KH ₂ PO ₄	1.35g
Esculin	1.0g
Acriflavin-HCl	12.5mg
Nalidixic acid.....	10.0mg
Ferric ammonium citrate supplement.....	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder and supplement from BD Diagnostic Systems.

Ferric Ammonium Citrate Supplement:**Composition per 10.0mL:**

Ferric ammonium citrate.....	0.5g

Preparation of Ferric Ammonium Citrate Supplement: Add ferric ammonium citrate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except ferric ammonium citrate supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile ferric ammonium citrate supplement. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Listeria* species from foods.

Deoxycholate Agar**Composition per liter:**

Agar.....	16.0g
Lactose	10.0g
NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
K ₂ HPO ₄	2.0g
Ferric citrate.....	1.0g
Sodium citrate.....	1.0g
Sodium deoxycholate.....	1.0g
Neutral Red.....	0.033g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the selective isolation, cultivation, enumeration, and differentiation of Gram-negative enteric microorganisms. *Escherichia coli* appears as large, flat, rose-red colonies. *Enterobacter* and *Klebsiella* species appear as large, mucoid, pale colonies with a pink center. *Proteus* and *Salmonella* species appear as large, colorless to tan colonies. *Shigella* species appear as colorless to pink colonies. *Pseudomonas* species appear as irregular colorless to brown colonies.

**Deoxycholate Agar
(Desoxycholate Agar)****Composition per liter:**

Agar.....	15.0g
Lactose	10.0g
Peptone	10.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.0g
Ferric citrate.....	1.0g
Sodium citrate.....	1.0g
Sodium deoxycholate.....	1.0g
Neutral Red.....	0.03g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid and BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes.

Use: For the selective isolation, cultivation, enumeration, and differentiation of Gram-negative enteric microorganisms. *Escherichia coli* appears as large, flat, rose-red colonies. *Enterobacter* and *Klebsiella* species appear as large, mucoid, pale colonies with a pink center. *Proteus* and *Salmonella* species appear as large, colorless to tan colonies. *Shigella* species appear as colorless to pink colonies. *Pseudomonas* species appear as irregular colorless to brown colonies.

Deoxycholate Citrate Agar**Composition per liter:**

Sodium citrate.....	50.0g
Agar.....	15.0g
Lactose	10.0g
Beef extract.....	5.0g
Peptone	5.0g
Na ₂ S ₂ O ₃ ·5H ₂ O.....	5.0g
Sodium deoxycholate.....	2.5g
Ferric citrate.....	1.0g
Neutral Red.....	0.025g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Dry the agar surface before use.

Use: For the selective isolation and cultivation of enteric pathogens, especially *Salmonella* and *Shigella* species.

Deoxycholate Citrate Agar

Composition per liter:

Sodium citrate	20.0g
Agar	17.0g
Lactose	10.0g
Meat, solids from infusion	10.0g
Peptic digest of animal tissue.....	10.0g
Sodium deoxycholate.....	5.0g
Ferric citrate.....	1.0g
Neutral Red.....	0.02g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Dry the agar surface before use.

Use: For the selective isolation and cultivation of enteric pathogens, especially *Salmonella* and *Shigella* species.

Deoxycholate Citrate Agar (Desoxycholate Citrate Agar)

Composition per liter:

Pork infusion.....	330.0g
Sodium citrate	20.0g
Agar	13.5g
Lactose	10.0g
Proteose peptone No. 3	10.0g
Sodium deoxycholate.....	5.0g
Ferric ammonium citrate.....	2.0g
Neutral Red.....	0.02g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Dry the agar surface before use.

Use: For the selective isolation and cultivation of enteric pathogens, especially *Salmonella* and *Shigella* species.

Deoxycholate Citrate Agar, Hynes

Composition per liter:

Agar	12.0g
Lactose	10.0g
Sodium citrate.....	8.5g
Na ₂ S ₂ O ₃ ·5H ₂ O	5.4g
Beef extract powder.....	5.0g
Peptone	5.0g
Sodium deoxycholate.....	5.0g
Ferric citrate.....	1.0g
Neutral Red.....	0.02g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Dry the agar surface before use.

Use: For the selective isolation, cultivation, and differentiation of enteric pathogens, especially *Salmonella* and *Shigella* species. Lactose-fermenting bacteria appear as pink colonies that may or may not be surrounded by a zone of precipitated deoxycholate. Nonlactose-fermenting bacteria appear as colorless colonies that are surrounded by a clear orange-yellow zone.

Deoxycholate Lactose Agar

Composition per liter:

Agar	15.0g
Lactose.....	10.0g
NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
Sodium citrate.....	2.0g
Sodium deoxycholate.....	0.5g
Neutral Red.....	0.033g

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Dry the agar surface before use.

Use: For the selective isolation, cultivation, and differentiation of enteric pathogens, especially *Salmonella*

and *Shigella* species. Lactose-fermenting bacteria appear as pink colonies that may or may not be surrounded by a zone of precipitated deoxycholate. Nonlactose-fermenting bacteria appear as colorless colonies that are surrounded by a clear orange-yellow zone. Used for the enumeration of coliform bacteria from milk and dairy products.

Dextrose Agar

Composition per liter:

Agar	15.0g
Glucose	10.0g
NaCl	5.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	3.0g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of microorganisms from foods.

Dextrose Agar

Composition per liter:

Agar	15.0g
Glucose	10.0g
Tryptose	10.0g
NaCl	5.0g
Beef extract	3.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a wide variety of microorganisms.

Dextrose Tryptone Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g

Glucose	5.0g
Bromcresol Purple	0.04g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of mesophilic and thermophilic aerobic microorganisms in food.

Dextrose Tryptone Agar

Composition per liter:

Agar	12.0g
Pancreatic digest of casein	10.0g
Glucose	5.0g
Bromcresol Purple	0.04g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and enumeration of “flat-sour” thermophiles and mesophiles in food. Acid-producing microorganisms such as “flat-sour” hemophiles appear as yellow colonies surrounded by a yellow zone.

Dextrose Tryptone Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Glucose	5.0g
Bromcresol Purple	0.04g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of “flat-sour” thermophiles and mesophiles in food. Acid-producing microorganisms such as “flat-sour” thermophiles turn the medium yellow.

Dextrose Tryptone Broth (m-Dextrose Tryptone Broth)

Composition per liter:

Pancreatic digest of casein	20.0g
Glucose	10.0g
Bromocresol Purple	0.04g
pH 6.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation, cultivation, and enumeration of “flat-sour” thermophiles and mesophiles in food by the membrane filter technique. Acid-producing microorganisms such as “flat-sour” thermophiles turn the medium yellow.

Dichloran 18% Glycerol Agar (DG18 Agar) (BAM M184)

Composition per liter:

Glycerol	220.0g
Agar	15.0g
Glucose	10.0g
Peptone	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Dichloran	2.0mg
Chloramphenicol	0.1g
pH 5.6 ± 0.2 at 25°C	

Preparation of Medium: Add components, except glycerol, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Add 220.0g of glycerol. Mix thoroughly. Bring volume to 1.0L with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. The final pH should be 5.6 and the final a_w should be 0.955. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: Used as a general purpose medium for the enumeration of molds from foods. For the enumeration and isolation of xerophilic molds from dried and semidried foods. This medium is preferred when the a_w of the analyzed food is 0.95 or lower. The low water activity of this medium reduces interference by bacteria and fast-growing fungi.

Dichloran-Glycerol Agar (DG18)

Composition per liter:

Glycerol	220.0g
Agar	15.0g

Glucose	10.0g
Peptone	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄	0.5g
Chloramphenicol	0.2g
Dichloran	2.0mg

pH 5.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except glycerol, to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Add 220.0g of glycerol. Mix thoroughly. Bring volume to 1.0L with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. The final pH should be 5.6. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: A selective low water activity (a_w) medium for xerophilic molds from dried and semi-dried foods (dried fruits, spices, confectionery, cereals, nuts, dried meat, and dried fish products) as well as a general purpose medium for counting yeasts and molds in foodstuffs.

Dichloran Rose Bengal Chloramphenicol Agar (DRBC Agar) (BAM M183)

Composition per liter:

Agar	15.0g
Glucose	10.0g
Peptone	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Chloramphenicol	0.1g
Dichloran solution	1.0mL
Rose Bengal solution	0.5mL
pH 5.6 ± 0.2 at 25°C	

Dichloran Solution:

Composition per 10.0mL:

Dichloran (2,6-dichloro-4-nitroaniline)	0.2.0g

Preparation of Dichloran Solution: Add dichloran to 10.0mL of distilled/deionized water. Mix thoroughly.

Rose Bengal Solution:

Composition per 10.0mL:

Rose Bengal	0.5g

Preparation of Rose Bengal Solution: Add Rose Bengal to 10.0mL of distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. The final pH should be 5.6. Temper in a water bath at 45°C. Pour into sterile Petri dishes or distribute into sterile tubes. This medium is light sensitive and should be stored in a dark, cool place until used. It is intended for spread plates only.

Use: For the analysis of food samples containing spreader molds, e.g., *Mucor* and *Rhizopus*. The dichloran and Rose Bengal slow down the growth of fast-growing fungi, thus allowing the detection of other fungi with slower growth rates.

Differential Broth for Lactic Streptococci

Composition per liter:

Sodium citrate	20.0g
Arginine	5.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
K ₂ HPO ₄	1.0g
Bromeresol Purple	0.02g
Skim milk (11% solution)	35.0mL

pH 6.2 ± 0.2 at 25°C

Preparation of Medium: Add components, except skim milk solution, to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Add 35.0mL of skim milk solution. Bring volume to 1.0L with distilled/deionized water. Place medium in a steam bath for 15 min. Cool to 25°C. Adjust pH to 6.2. Distribute 7.0mL volumes into screw-capped tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C. Allow autoclave to cool below 70°C before opening door.

Use: For the cultivation and differentiation of *Lactobacillus lactis*, *Lactobacillus lactis* subspecies *cremoris*, and *Lactobacillus lactis* subspecies *diacetylactis*. Lactose-fermenting bacteria such as *Lactobacillus lactis* subspecies *cremoris* turn the medium yellow. Arginine-utilizing bacteria such as *Lactobacillus lactis* initially turn the medium yellow but then turn back to violet. Citrate-utilizing bacteria such as *Lactobacillus lactis* subspecies *diacetylactis* turn the medium violet and produce CO₂ that is trapped as a bubble in the Durham tube.

DNase Agar

Composition per liter:

Tryptose	20.0g
Agar	12.0g
NaCl	5.0g
Deoxyribonucleic acid	2.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms, especially *Staphylococcus* species and *Serratia marcescens*, based on their production of deoxyribonuclease.

DNase Test Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	15.0g
NaCl	5.0g
Papaic digest of soybean meal	5.0g
Deoxyribonucleic acid	2.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms, especially *Staphylococcus* species and *Serratia marcescens*, based on their production of deoxyribonuclease.

DNase Test Agar with Methyl Green

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Deoxyribonucleic acid	2.0g
Methyl Green	0.05g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms, especially *Staphylococcus* species and *Serratia marcescens*, based on their production of deoxyribonuclease.

DNase Test Agar with Toluidine Blue

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Deoxyribonucleic acid	2.0g
Toluidine Blue	0.1g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms, especially *Staphylococcus* species and *Serratia marcescens*, based on their production of deoxyribonuclease.

Dorset Egg Medium

Composition per liter:

Homogenized whole egg	950.0mL
Glycerol	50.0mL

pH 6.8–7.4 at 25°C

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18–24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Filter sterilize glycerol. Combine glycerol and homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes.

Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the maintenance of *Mycobacterium* species.

Double-Strength Crude *Lactobacillus* Medium

Composition per 1025.0mL:

Yeast extract (Basamine)	20.0g
Sucrose	20.0g
Casein hydrolysate	15.0g
Potassium acetate	3.0g
Histidine-HCl·H ₂ O	2.0g
Ascorbic acid	1.0g
Pyridoxamine-HCl	33.0µg
Salts solution A	20.0mL
Salts solution B	5.0mL

pH 5.4 ± 0.2 at 25°C

Salts Solution A:

Composition per liter:

K ₂ HPO ₄ ·3H ₂ O	16.5g
KH ₂ PO ₄ ·H ₂ O	16.5g

Preparation of Salts A: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Salts Solution B:

Composition per liter:

MgSO ₄ ·7H ₂ O	8.0g
FeSO ₄ ·7H ₂ O	0.4g
NaCl	0.4g
HCl, concentrated	0.1mL
MnSO ₄ ·H ₂ O	0.1mL

Preparation of Salts B: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except salts solution A and salts solution B, to distilled/deionized water and bring volume to 975.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 20.0mL of sterile salts solution A and 5.0mL of sterile salts solution B. Mix thoroughly. Adjust pH to 5.4. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Lactobacillus* species.

Double-Strength Crude Medium for *Lactobacillus*

Composition per liter:

Sucrose	20.0g
Yeast extract	20.0g

Casein hydrolysate.....	15.0g
Potassium acetate.....	3.0g
Histidine-HCl.....	2.0g
Ascorbic acid.....	1.0g
Pyridoxamine-HCl.....	33.0µg
Salts A.....	20.0mL
Salts B.....	5.0mL

pH 5.4 ± 0.2 at 25°C

Salts A:**Composition** per 100.0mL:

KH ₂ PO ₄ ·H ₂ O.....	16.5g
K ₂ HPO ₄ ·3H ₂ O.....	16.5g

Preparation of Salts A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Salts B:**Composition** per 100.0mL:

MgSO ₄ ·7H ₂ O.....	8.0g
FeSO ₄ ·7H ₂ O.....	0.4g
MnSO ₄ ·H ₂ O.....	0.4g
NaCl.....	0.4g
HCl, concentrated.....	0.1mL

Preparation of Salts B: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 5.4 with acetic acid. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus* species.

Doyle and Roman Enrichment Medium**Composition** per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
NaCl.....	5.0g
Sodium succinate.....	3.0g
Yeast extract.....	2.0g
Glucose.....	1.0g
NaHSO ₃	0.1g
L-Cysteine-HCl·H ₂ O.....	0.1g
Horse blood, lysed.....	70.0mL
Antibiotic solution.....	10.0mL

pH 7.0 ± 0.2 at 25°C

Antibiotic Solution:**Composition** per 10.0mL:

Cycloheximide.....	0.05g
Vancomycin.....	0.015g
Trimethoprim lactate.....	5.0mg
Polymyxin B.....	200,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution and horse blood, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution and horse blood. Mix thoroughly. Aseptically distribute into sterile flasks in 90.0–100.0mL volumes.

Use: For the cultivation and enrichment of *Campylobacter* species from foods.

DRBC Agar**(Dichloran Rose Bengal Chloramphenicol Agar)****Composition** per liter:

Agar.....	15.0g
Glucose.....	10.0g
Peptone.....	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
Rose Bengal.....	0.025g
Dichloran.....	0.002g
Chloramphenicol solution.....	10.0mL

pH 5.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Chloramphenicol Solution:**Composition** per 10.0mL:

Chloramphenicol.....	0.1g

Preparation of Chloramphenicol Solution:

Add chloramphenicol to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except chloramphenicol solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile chloramphenicol solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and enumeration of yeasts and molds associated with food spoilage.

**Duncan-Strong
Sporulation Medium, Modified
(DS Sporulation Medium, Modified)
(Sporulation Medium, Modified)
(BAM M45)**

Composition per liter:

Proteose peptone	15.0g
Na ₂ HPO ₄ ·7H ₂ O	10.0g
Raffinose	4.0g
Yeast extract	4.0g
Sodium thioglycolate	1.0g
pH 7.8 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Adjust pH to 7.8 with filter-sterilized 0.66M Na₂CO₃. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and induction of sporulation of *Clostridium perfringens*.

**E Agar
(m-E Agar)**

Composition per liter:

Yeast extract	30.0g
Agar	15.0g
NaCl	15.0g
Pancreatic digest of gelatin	10.0g
Esculin	1.0g
Nalidixic acid	0.25g
NaN ₃	0.15g
Cycloheximide	0.05g
TTC solution	15.0mL
pH 7.1 ± 0.2 at 25°C	

TTC Solution:

Composition per 15.0mL:

2,3,5-Triphenyltetrazolium chloride	0.15g

Preparation of TTC Solution: Add triphenyltetrazolium chloride to distilled/deionized water and bring volume to 15.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except TTC solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile TTC solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Use: For the isolation, cultivation, and enumeration of enterococci in water by the membrane filter method. It is used in conjunction with esculin iron agar.

**Eagle's Minimal Essential Medium
with Earle's Salts
and Nonessential Amino Acids
(MEM with Earle's Salts
and Nonessential Amino Acids)
(BAM M46)**

Composition per liter:

NaCl	6.8g
NaHCO ₃	2.2g
Glucose	1.0g
KCl	0.4g
CaCl ₂ ·2H ₂ O	0.265g
MgSO ₄ ·7H ₂ O	0.2g
L-Arginine·H ₂ O	0.15g
NaH ₂ PO ₄ ·H ₂ O	0.14g
L-Arginine-HCl	0.126g
L-Lysine-HCl	72.5mg
L-Tyrosine, disodium salt	52.1mg
L-Leucine	52.0mg
L-Threonine	48.0mg
L-Valine	46.0mg
L-Histidine-HCl·H ₂ O	42.0mg
D-Phenylalanine	32.0mg
L-Cysteine·2HCl	31.29mg
L-Methionine	15.0mg
L-Glutamic acid	14.7mg
L-Aspartic acid	13.3mg
L-Proline	11.5mg
L-Serine	10.5mg
L-Tryptophan	10.0mg
L-Alanine	8.9mg
Phenol Red	10.0mg
L-Glycine	7.5mg
<i>D</i> -Inositol	2.0mg
D-Calcium pantothenate	1.0mg
Choline chloride	1.0mg
Folic acid	1.0mg
Nicotinamide	1.0mg
Pyridoxal-HCl	1.0mg
Thiamine-HCl	1.0mg
Riboflavin	0.1mg

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to 1.0L of distilled/deionized water. Mix thoroughly. Filter sterilize.

Use: For the cultivation of animal cells in tissue culture, for example cells for viral detection and identification by characteristic cytopathic effects.

EB Motility Medium (BAM M48)

Composition per liter:

Peptone or gelysate	10.0g
NaCl	5.0g
Agar	4.0g
Beef extract	3.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling for 1–2 min. Distribute into tubes in 8.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on motility.

EC Broth (*Escherichia coli* Broth) (EC Medium) (BAM M49)

Composition per liter:

Pancreatic digest of casein	20.0g
Lactose	5.0g
NaCl	5.0g
K ₂ HPO ₄	4.0g
Bile salts mixture	1.5g
KH ₂ PO ₄	1.5g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 12 min at 15 psi pressure–121°C. Cool broth as quickly as possible.

Use: For the cultivation and differentiation of coliform bacteria at 37°C and of *Escherichia coli* at 45.5°C.

EC Broth (*Escherichia coli* Broth) (EC Medium) (BAM M49)

Composition per liter:

Pancreatic digest of casein	20.0g
Lactose	5.0g
NaCl	5.0g
K ₂ HPO ₄	4.0g
KH ₂ PO ₄	1.5g

Bile salts No.3	1.12g
Novobiocin solution	10.0mL
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Novobiocin Solution:

Composition per 50.0mL:

Novobiocin	0.1g

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL novobiocin solution. Mix thoroughly. Aseptically distribute to sterile tubes or flasks.

Use: A selective enrichment broth for the growth of *E. coli* O157, from food and environmental samples.

EC Broth with MUG (BAM M50)

Composition per liter:

Pancreatic digest of casein	20.0g
Lactose	5.0g
NaCl	5.0g
K ₂ HPO ₄	4.0g
Bile salts mixture	1.5g
KH ₂ PO ₄	1.5g
4-Methylumbelliferyl-β-D-glucuronide (MUG)	0.05g
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the detection of *Escherichia coli* in food samples by a fluorogenic procedure.

EC Medium, Modified with Novobiocin

Composition per liter:

Tryptone	20.0g
NaCl	5.0g
Lactose	5.0g
K ₂ HPO ₄	4.0g
KH ₂ PO ₄	1.5g

Bile salts.....	1.12g
Novobiocin supplement.....	10.0mL
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder and supplement from BD Diagnostic Systems.

Novobiocin Supplement:

Composition per 10.0mL:

Sodium novobiocin.....	20.0mg

Preparation of Novobiocin Supplement: Add Sodium novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile novobiocin supplement. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Escherichia coli* O157:H7.

ECD Agar, Fluorocult (Fluorocult ECD Agar)

Composition per liter:

Peptone from casein.....	20.0g
Agar.....	15.0g
NaCl.....	5.0g
Lactose.....	5.0g
K ₂ HPO ₄	4.0g
Bile salt mixture.....	1.5g
KH ₂ PO ₄	1.5g
Tryptophan.....	1.0g
4-Methylumbelliferyl-β-D-glucuronide.....	0.07g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes. The prepared broth is clear and yellowish brown.

Use: For the detection of *E. coli* in meats. The medium complies with the German-DIN-Norm 10110 for the examination of meat, with the regulations according to § 35 LMBG (06.00/36) for the examination of food and with ISO Standard 6391 (1996) for the enumeration of *E. coli* in meat and meat products. The bile salt mixture of this medium largely inhibits the accompanying flora not usually found in the intestines. Using fluorescence under UV light and a posi-

tive indole reaction, *E. coli* colonies can be identified among the grown colonies.

EE Broth

(Enterobacteriaceae Enrichment Broth)

Composition per liter:

Ox bile.....	20.0g
Peptone.....	10.0g
Na ₂ HPO ₄	6.45g
Glucose.....	5.0g
KH ₂ PO ₄	2.0g
Brilliant Green.....	0.0135g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into flasks in 100.0mL volumes. Gently heat at 100°C for 30 min. Do not autoclave. Cool rapidly to 25°C.

Use: For the cultivation and enrichment of members of the Enterobacteriaceae in the examination of foods and animal feed. Used in conjunction with tryptone soy broth. Bacteria belonging to the Enterobacteriaceae turn this medium turbid and yellow-green.

EE Broth, Mossel

(Enterobacteriaceae

Enrichment Broth, Mossel)

Composition per liter:

Enzymatic hydrolysate of protein.....	10.0g
Na ₂ HPO ₄	8.0g
Glucose.....	5.0g
KH ₂ PO ₄	2.0g
Oxgall.....	0.1g
Brilliant Green.....	0.0135g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into flasks in 120.0mL volumes. Gently heat at 100°C for 30 min. Do not autoclave. Cool rapidly to 25°C.

Use: For the cultivation and enrichment of members of the Enterobacteriaceae in the examination of foods and animal feed. Used in conjunction with tryptone soy broth. Bacteria belonging to the Enterobacteriaceae turn this medium turbid and yellow-green.

Egg Yolk Agar**Composition per liter:**

Proteose peptone No. 2	40.0g
Agar	25.0g
Na ₂ HPO ₄	5.0g
Glucose	2.0g
NaCl	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Egg yolk emulsion	100.0mL
Hemin solution	1.0mL

pH 7.6 ± 0.2 at 25°C

Hemin Solution:**Composition per 100.0mL:**

Hemin	0.5g
NaOH (1N solution)	20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Egg Yolk Emulsion:**Composition:**

Chicken egg yolks	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and differentiation of *Clostridium* species and some other anaerobic bacteria.

Egg Yolk Agar, Modified**Composition per liter:**

Agar	20.0g
Pancreatic digest of casein	15.0g
Vitamin K ₁	10.0g
NaCl	5.0g
Papaic digest of soybean meal	5.0g
Yeast extract	5.0g
L-Cystine	0.4g
Hemin	5.0mg
Egg yolk emulsion	100.0mL

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Egg Yolk Emulsion:**Composition:**

Chicken egg yolks	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and differentiation of *Clostridium* species and some other anaerobic bacteria.

Egg Yolk Emulsion**Composition per 100mL:**

Sterile saline	70.0mL
Egg yolk	30.0mL

Source: Sterile egg yolk emulsion is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min or soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites, placing egg yolks into a sterile container. Use enough eggs to produce at least 30.0mL egg yolk. Homogenize by shaking. Add 0.9g NaCl to distilled/deionized water and bring volume to 100.0mL. Sterilize the saline solution by filtration or by autoclaving for 15 min at 15 psi pressure–121°C. If autoclaving is used, cool to 25°C. Aseptically add 30.0mL homogenized egg yolks to 70.0mL of sterile saline solution. Mix thoroughly.

Use: Sterile stabilized emulsion of egg yolk recommended for use in various culture media.

Egg Yolk Emulsion**Composition per 100mL:**

NaCl	0.45g
Egg yolk	50.0mL

Preparation of Medium: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. or soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites, placing egg yolks into a sterile container. Use enough eggs to produce at least 50.0mL egg yolk. Homogenize by shaking. Add 0.45g NaCl to distilled/deionized water and bring volume to 50.0mL. Sterilize the saline solution by filtration or by autoclaving for 15 min at 15 psi pressure–121°C. If autoclaving is used, cool to 25°C. Aseptically add 50.0mL homogenized egg yolks to 50.0mL of the sterile NaCl solution. Mix thoroughly.

Use: Sterile stabilized emulsion of egg yolk recommended for use in various culture media.

Egg Yolk Emulsion, 50% (BAM M51)

Composition per 100.0mL:

Chicken egg yolks.....variable
NaCl (0.85% solution) 40.0mL

Preparation of Egg Yolk Emulsion: Wash fresh eggs with stiff brush and drain. Soak eggs in 70% ethanol for 1 hr. Crack eggs aseptically and separate yolks from whites. Remove egg yolks with a sterile syringe or a wide-mouth pipet. Place 50.0mL of egg yolks into a sterile container. Add 50.0mL sterile 0.85% saline.

Use: For use in media requiring egg yolk emulsion.

Egg-Yolk Tellurite Emulsion 20%

Composition per 100mL:

NaCl.....0.425g
K₂TeO₃.....0.21g
Egg yolk..... 20.0mL

Preparation of Medium: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. or soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites, placing egg yolks into a sterile container. Use enough eggs to produce at least 20.0mL egg yolk. Homogenize by shaking. Add 0.45g NaCl and 0.21g K₂TeO₃ to distilled/deionized water and bring volume to 80.0mL. Sterilize the saline-tellurite solution by filtration or by autoclaving for 15 min at 15 psi pressure–121°C. If autoclaving is used, cool to 25°C. Aseptically add 20.0mL homogenized egg yolks to

80.0mL of the sterile saline-tellurite solution. Mix thoroughly.

Use: For use in various culture media. It may be added directly to nutrient media for the identification of *Clostridium*, *Bacillus* and *Staphylococcus* species by their lipase activity.

Eijkman Lactose Medium

Composition per liter:

Pancreatic digest of casein..... 15.0g
K₂HPO₄..... 10.0g
KH₂PO₄..... 4.0g
Lactose..... 3.0g
NaCl..... 2.5g

pH 6.8 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Escherichia coli* from other coliform organisms based on their ability to ferment lactose and produce gas.

Eijkman Lactose Medium

Composition per liter:

Tryptose..... 15.0g
NaCl..... 5.0g
K₂HPO₄..... 4.0g
Lactose..... 3.0g
KH₂PO₄..... 1.5g

pH 6.8 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Escherichia coli* from other coliform organisms based on their ability to ferment lactose and produce gas.

Elliker Agar

Composition per liter:

Pancreatic digest of casein..... 20.0g
Agar..... 15.0g
Glucose..... 5.0g
Lactose..... 5.0g
Sucrose..... 5.0g
Yeast extract..... 5.0g
NaCl..... 4.0g
Gelatin..... 2.5g

Sodium acetate.....	1.5g
Ascorbic acid	0.5g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of lactic streptococci.

Elliker Agar

Composition per liter:

Pancreatic digest of casein.....	20.0g
Agar	15.0g
Yeast extract.....	10.0g
Gelatin.....	4.0g
Glucose	3.0g
Ascorbic acid	2.5g
Lactose	2.5g
NaCl.....	2.5g
Sodium acetate.....	2.5g
Sucrose.....	2.5g
pH 6.8 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of streptococci and lactobacilli of importance in the dairy industry.

Elliker Broth

Composition per liter:

Pancreatic digest of casein.....	20.0g
Yeast extract.....	10.0g
Gelatin.....	4.0g
Glucose	3.0g
Ascorbic acid	2.5g
Lactose	2.5g
NaCl.....	2.5g
Sodium acetate.....	2.5g
Sucrose.....	2.5g
pH 6.8 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of streptococci and lactobacilli of importance in the dairy industry.

Elliker Broth

Composition per liter:

Pancreatic digest of casein.....	20.0g
Dextrose.....	5.0g
Lactose.....	5.0g
Sucrose.....	5.0g
Yeast extract.....	5.0g
NaCl.....	4.0g
Gelatin.....	2.5g
Sodium acetate.....	1.5g
Ascorbic acid	0.5g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of streptococci and lactobacilli of importance in the dairy industry.

Elliker Lactose Broth (ATCC Medium 2333)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose.....	10.0g
Yeast extract.....	5.0g
NaCl.....	4.0g
Gelatin.....	2.5g
Ascorbic acid	2.5g
Sodium acetate.....	1.5g
pH 7.0 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of streptococci and lactobacilli of importance in the dairy industry.

EMB Agar (Eosin Methylene Blue Agar)

Composition per liter:

Agar	13.5g
Pancreatic digest of casein.....	10.0g
Lactose.....	5.0g
Sucrose.....	5.0g
K ₂ HPO ₄	2.0g

Eosin Y	0.4g
Methylene Blue	0.065g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and differentiation of Gram-negative enteric bacteria based on lactose fermentation. Bacteria that ferment lactose, especially the coliform bacterium *Escherichia coli*, appear as colonies with a green metallic sheen or blue-black to brown color. Bacteria that do not ferment lactose appear as colorless or transparent, light purple colonies.

EMB Agar Base

Composition per liter:

Agar	15.0g
Peptone	10.0g
K ₂ HPO ₄	2.0g
Eosin Y	0.4g
Methylene Blue	0.065g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and differentiation of Gram-negative enteric bacteria based on lactose fermentation. Bacteria that ferment lactose, especially the coliform bacterium *Escherichia coli*, appear as colonies with a green metallic sheen or blue-black to brown color. Bacteria that do not ferment lactose appear as colorless or transparent, light purple colonies.

EMB Agar, Modified

(Eosin Methylene Blue Agar, Modified)

Composition per liter:

Agar	15.0g
Lactose	10.0g
Pancreatic digest of gelatin	10.0g
K ₂ HPO ₄	2.0g

Eosin Y	0.4g
Methylene Blue	0.065g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 60°C. Shake medium to oxidize methylene blue. Pour into sterile Petri dishes. Swirl flask while pouring plates to distribute precipitate.

Use: For the isolation, cultivation, and differentiation of Gram-negative enteric bacteria based on lactose fermentation. Bacteria that ferment lactose, especially the coliform bacterium *Escherichia coli*, appear as colonies with a green metallic sheen or blue-black to brown color. Bacteria that do not ferment lactose appear as colorless or transparent, light purple colonies.

Endo Agar

Composition per liter:

Agar	15.0g
Lactose	10.0g
Peptic digest of animal tissue	10.0g
K ₂ HPO ₄	3.5g
Na ₂ SO ₃	2.5g
Basic Fuchsin	0.5g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Basic Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes. Swirl flask while pouring plates to keep precipitate in suspension. Protect from the light.

Use: For the selective isolation, cultivation, and differentiation of coliform and other enteric microorganisms based on their ability to ferment lactose. Lactose-fermenting bacteria appear as dark red colonies with a gold metallic sheen. Lactose-nonfermenting bacteria appear as colorless or translucent colonies.

Endo Agar**Composition per liter:**

Agar	10.0g
Lactose	10.0g
Peptic digest of animal tissue.....	10.0g
K ₂ HPO ₄	3.5g
Na ₂ SO ₃	2.5g
Basic Fuchsin solution	4.0mL
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Caution: Basic Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Basic Fuchsin Solution:**Composition per 10.0mL:**

Basic Fuchsin.....	1.0g
Ethanol (95% solution)	10.0mL

Preparation of Basic Fuchsin Solution: Add Basic Fuchsin to 10.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes. Swirl flask while pouring plates to keep precipitate in suspension. Protect from the light.

Use: For the selective isolation, cultivation, and differentiation of coliform and other enteric microorganisms based on their ability to ferment lactose. Lactose-fermenting bacteria appear as dark red colonies with a gold metallic sheen. Lactose-nonfermenting bacteria appear as colorless or translucent colonies.

Endo Agar, LES**(m-Endo Agar, LES)****Composition per liter:**

Agar	10.0g
Lactose	9.4g
Tryptose	7.5g
NaCl.....	3.7g
Peptone.....	3.7g
Pancreatic digest of casein.....	3.7g
K ₂ HPO ₄	3.3g
Na ₂ SO ₃	1.6g
Yeast extract.....	1.2g
KH ₂ PO ₄	1.0g
Sodium deoxycholate.....	0.1g
Sodium lauryl sulfate.....	0.05g
Basic Fuchsin solution	8.0mL
pH 7.2 ± 0.2 at 25°C	

Caution: Basic Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Basic Fuchsin Solution:**Composition per 10.0mL:**

Basic Fuchsin.....	1.0g
Ethanol (95% solution)	10.0mL

Preparation of Basic Fuchsin Solution: Add Basic Fuchsin to 10.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes. Swirl flask while pouring plates to keep precipitate in suspension. Protect from the light.

Use: For the cultivation and enumeration of coliform bacteria from water by the membrane filter method.

Endo Broth**(m-Endo Broth)****Composition per liter:**

Lactose	12.5g
Peptone	10.0g
NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
K ₂ HPO ₄	4.375g
Na ₂ SO ₃	2.1g
Yeast extract.....	1.5g
KH ₂ PO ₄	1.375g
Basic Fuchsin.....	1.05g
Sodium deoxycholate.....	0.1g
Ethanol (95% solution)	20.0mL
pH 7.2 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Basic Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Preparation of Medium: Add ethanol to approximately 900.0mL of distilled/deionized water. Add remaining components. Bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Rapidly cool broth below 45°C. Do not autoclave. Use 1.8–2.0mL for each filter pad. Protect from the light. Prepare broth freshly.

Use: For the cultivation and enumeration of coliform bacteria from water by the membrane filter method.

Enriched Nutrient Agar**Composition per liter:**

Agar	15.0g
Heart, solids from infusion	12.5g
Yeast extract	3.3g
Peptone	2.1g
NaCl	2.1g
Beef extract	0.42g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Micrococcus luteus*.

Enriched Nutrient Broth**Composition per liter:**

Beef heart, infusion from	250.0g
Tryptose	5.0g
Pancreatic digest of gelatin	3.38g
NaCl	2.5g
Yeast extract	2.5g
Beef extract	2.02g

pH 7.2 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus licheniformis*, *Bacillus polymyxa*, *Bacillus subtilis*, *Escherichia coli*, *Listonella anguillarum*, *Micrococcus luteus*, *Pseudomonas aeruginosa*, *Salmonella choleraesuis*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus* species, and *Vibrio cholerae*.

Enriched Nutrient Broth**Composition per liter:**

Beef heart, infusion from	300.0g
Tryptose	7.5g
NaCl	3.0g
Yeast extract	3.0g
Peptone	2.5g
NaCl	2.5g
Beef extract	0.5g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fastidious bacteria.

**Enrichment Broth
for *Aeromonas hydrophila*****Composition per liter:**

NaCl	5.0g
Maltose	3.5g
Yeast extract	3.0g
Bile salts No. 3	1.0g
L-Cysteine-HCl-H ₂ O	0.3g
Bromthymol Blue	0.03g
Novobiocin	5.0mg

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and enrichment of *Aeromonas hydrophila*.

Enrichment Broth, pH 7.3**Composition per liter:**

Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g
Yeast extract	6.0g
Nalidixic acid solution	8.0mL
Cycloheximide solution	5.1mL
Acriflavin-HCl solution	3.0mL

pH 7.3 ± 0.2 at 25°C

Cycloheximide Solution:**Composition per 10.0mL:**

Cycloheximide	0.1g
Ethanol, absolute	4.0mL

Preparation of Cycloheximide Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:**Composition per 10.0mL:**

Nalidixic acid	0.05g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin-HCl Solution:**Composition per 10.0mL:**

Acriflavin-HCl	0.05g

Preparation of Acriflavin-HCl Solution: Add acriflavin-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except cycloheximide solution, nalidixic acid solution, and acriflavin-HCl solution—to distilled/deionized water and bring volume to 983.9mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.1mL of sterile cycloheximide solution, 8.0mL of sterile nalidixic acid solution, and 3.0mL of sterile acriflavin-HCl solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the isolation, cultivation, and enrichment of a variety of microorganisms from nondairy foods.

Enrichment Broth, pH 7.3

Composition per liter:

Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g
Yeast extract	6.0g
Nalidixic acid solution	8.0mL
Cycloheximide solution	5.1mL
Acriflavin-HCl solution	2.0mL
pH 7.3 ± 0.2 at 25°C	

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.1g
Ethanol, absolute	4.0mL

Preparation of Cycloheximide Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid	0.05g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin-HCl Solution:

Composition per 10.0mL:

Acriflavin-HCl	0.05g

Preparation of Acriflavin-HCl Solution: Add acriflavin-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except cycloheximide solution, nalidixic acid solution, and acriflavin-HCl solution—to distilled/deionized

water and bring volume to 984.9mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.1mL of cycloheximide solution, 8.0mL of nalidixic acid solution, and 2.0mL of acriflavin-HCl solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the isolation, cultivation, and enrichment of a variety of microorganisms from milk and dairy products.

Enrichment Broth, pH 7.3 with Pyruvate

Composition per liter:

Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g
Yeast extract	6.0g
Pyruvate solution	11.1mL
Nalidixic acid solution	8.0mL
Cycloheximide solution	5.1mL
Acriflavin-HCl solution	3.0mL
pH 7.3 ± 0.2 at 25°C	

Pyruvate Solution:

Composition per 20.0mL:

Sodium pyruvate	2.0g

Preparation of Pyruvate Solution: Add sodium pyruvate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.1g
Ethanol, absolute	4.0mL

Preparation of Cycloheximide Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid	0.05g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin-HCl Solution:

Composition per 10.0mL:

Acriflavin-HCl	0.05g

Preparation of Acriflavin-HCl Solution: Add acriflavin-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except pyruvate solution, cycloheximide solution, nalidixic acid solution, and acriflavin-HCl solution—to distilled/deionized water and bring volume to 972.8mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 45°–50°C. Aseptically add 11.1mL of sterile pyruvate solution. Mix thoroughly. Inoculate medium and incubate at 30°C for 6 hr. Aseptically add 5.1mL of sterile cycloheximide solution, 8.0mL of sterile nalidixic acid solution, and 3.0mL of sterile acriflavin-HCl solution. Mix thoroughly.

Use: For the isolation, cultivation, and enrichment of a *Listeria* species from nondairy foods.

Enrichment Broth, pH 7.3 with Pyruvate Composition per liter:

Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g
Yeast extract	6.0g
Pyruvate solution	11.1mL
Nalidixic acid solution	8.0mL
Cycloheximide solution	5.1mL
Acriflavin-HCl solution	2.0mL
pH 7.3 ± 0.2 at 25°C	

Pyruvate Solution:

Composition per 20.0mL:

Sodium pyruvate	2.0g

Preparation of Pyruvate Solution: Add sodium pyruvate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.1g
Ethanol, absolute	4.0mL

Preparation of Cycloheximide Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid	0.05g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin-HCl Solution:

Composition per 10.0mL:

Acriflavin-HCl	0.05g

Preparation of Acriflavin-HCl Solution: Add acriflavin-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except pyruvate solution, cycloheximide solution, nalidixic acid solution, and acriflavin-HCl solution—to distilled/deionized water and bring volume to 973.8mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 45°–50°C. Aseptically add 11.1mL of sterile pyruvate solution. Mix thoroughly. Inoculate medium and incubate at 30°C for 6 hr. Aseptically add 5.1mL of sterile cycloheximide solution, 8.0mL of sterile nalidixic acid solution, and 2.0mL of sterile acriflavin-HCl solution. Mix thoroughly.

Use: For the isolation, cultivation, and enrichment of a *Listeria* species from milk and dairy products.

Entamoeba dispar Axenic Culture Medium

Composition per liter:

YI-S solution	990.0mL
Gastric mucin stock suspension	10.0mL

YI-S Solution:

Composition per liter:

YI broth	880.0mL
Bovine serum, heat inactivated	100.0mL
Vitamin mixture 18	20.0mL

Source: Vitamin mixture 18 is available from Biofluids, Inc., Rockville, MD.

YI Broth:

Composition per liter:

YI base stock	780.0mL
10× Glucose buffer stock	100.0mL

YI Base Stock:

Composition per 780.0mL:

Yeast extract	30.0g
L-Cysteine-HCl	1.0g
NaCl	1.0g
Ascorbic acid	0.2g
Ferric ammonium citrate	228.0mg

10× Glucose Buffer Stock:

Composition per 100.0.0mL:

Glucose	10.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	0.6g

Preparation of 10× Glucose Buffer Stock:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of YI Base Stock: Add components to 600.0mL of distilled/deionized water. Mix thoroughly. Bring volume to 780.0mL with distilled/deionized water. Adjust pH to 6.8 with 1N NaOH. Distribute in 78.0mL aliquots to 100.0mL screw-capped bottles. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature.

Preparation of YI Broth: Aseptically add 10.0mL of 10× glucose buffer stock to 78.0mL of cooled YI base stock. Adjust osmolarity with NaCl to 380.0milliosmols/kg

Preparation of YI-S Solution: Aseptically add 2.0mL of vitamin mixture 18 and 10.0mL of heat-inactivated bovine serum to 88.0mL of YI broth. Distribute in 12.0mL aliquots to 16 x 125mm screw-capped test tubes. Store at 4°C in the dark with the caps screwed on tightly. Use within 96 hr.

Gastric Mucin Stock Suspension:

Composition per 100.0mL:

Gastric mucin 1.0g

Preparation of Gastric Mucin Stock Suspension: Add gastric mucin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Aseptically add 0.12mL of gastric mucin stock suspension (thoroughly mix by swirling before use) to 12.0mL of the complete YI-S solution in a 16 x 125mm screw-capped test tube. Store at 4°C in the dark with the caps screwed on tightly. Use within 96 hr.

Use: For the cultivation of *Entamoeba dispar*.

***Entamoeba Medium* (Endamoeba Medium)**

Composition per liter:

Liver infusion 272.0g
Rice powder 14.2g
Agar 11.0g
Proteose peptone 5.5g
Sodium glycerophosphate 3.0g
NaCl 2.7g
Horse serum 50.0mL
pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systemss.

Rice Powder:

Composition per 15.0g:

Rice powder 15.0g

Preparation of Rice Powder: Sterilize rice powder at 160°C for 60 min. Do not overheat or rice powder will scorch.

Preparation of Medium: Add components, except horse serum and rice powder, to distilled/deionized water and bring volume to 994.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 7.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position. Aseptically add enough sterile horse serum to each tube to cover about half the slant. Aseptically add 0.1g of sterile rice powder to each tube.

Use: For the cultivation of *Entamoeba histolytica*.

Enteric Fermentation Base (Fermentation Base for *Campylobacter*)

Composition per liter:

Peptic digest of animal tissue 10.0g
NaCl 5.0g
Beef extract 3.0g
Carbohydrate solution 100.0mL
Andrade's indicator 10.0mL
pH 7.2 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate 10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Glucose, lactose, mannitol, sucrose, adonitol, arabinose, cellobiose, dulcitol, glycerol, inositol, salicin, xylose, or other carbohydrates may be used. For the preparation of expensive carbohydrate solutions (adonitol, arabinose, cellobiose, dulcitol, glycerol, inositol, salicin, or xylose), 5.0g of carbohydrate per 100.0mL of distilled/deionized water may be used.

Andrade's Indicator:

Composition per 100.0mL:

NaOH (1N solution) 16.0mL
Acid Fuchsin 0.1g

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Preparation of Andrade's Indicator: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes that contain an inverted Durham tube in 9.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 1.0mL of sterile carbohydrate solution per tube. Mix thoroughly.

Use: For the cultivation and differentiation of a variety of bacteria based on their ability to ferment different carbohydrates. Bacteria that produce acid from carbohydrate fermentation turn the medium dark pink to red. Bacteria that produce gas have a bubble trapped in the Durham tube.

Enterobacter Medium

Composition per 800.0mL:

Casein hydrolysate.....	2.0g
K ₂ HPO ₄	1.4g
K ₂ SO ₄	1.0g
Yeast extract.....	1.0g
KH ₂ PO ₄	0.6g
MgSO ₄	0.5g
Glycerol.....	20.0mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Enterobacter* species and *Klebsiella pneumoniae*.

Enterococci Broth, Chromocult (Chromocult Enterococci Broth)

Composition per liter:

Peptone.....	8.6g
NaCl.....	6.4g
Tween 80.....	2.2g
NaN ₃	0.6g
5-Bromo-4-chloro-3-indolyl-β-D-glucopyranoside (X-GLU).....	0.04

pH 7.5 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix well. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. The prepared broth is clear and yellowish.

Use: For the detection of enterococci in food and water. The sodium-azide present in this medium largely inhibits the growth of the accompanying, and

especially the Gram-negative microbial flora while sparing the enterococci. The reaction occurs because the substrate X-GLU (5-bromo-4-chloro-3-indolyl-β-D-glucopyranoside) is cleaved, stimulated by selected peptones, by the enzyme β-D-glucosidase which is characteristic for enterococci. This results in an intensive blue-green color of the broth. Azide, at the same time, prevents a false positive result by most other β-D-glucosidase positive bacteria. Therefore, the color change of the broth largely confirms the presence of enterococci and group D-streptococci.

Enterococci Confirmatory Agar

Composition per liter:

Agar.....	15.0g
Glucose.....	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	5.0g
NaN ₃	0.4g
Methylene Blue.....	10.0mg

pH 8.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position. Add sufficient amount of Enterococci confirmatory broth to cover half the slant.

Use: For the identification of enterococci from water by the confirmatory test.

Enterococci Confirmatory Broth

Composition per liter:

NaCl.....	65.0g
Glucose.....	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	5.0g
NaN ₃	0.4g
Methylene Blue.....	10.0mg
Penicillin.....	650U

pH 8.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except penicillin, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add penicillin. Mix thoroughly. Add sufficient amount of Enterococci confirmatory broth to cover half the slant of Enterococci confirmatory agar.

Use: For the identification of enterococci from water by the confirmatory test.

Enterococci Presumptive Broth

Composition per liter:

Glucose	5.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
NaN ₃	0.4g
Bromthymol Blue	32.0mg
pH 8.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and identification of enterococci from water by the presumptive test. Bacteria that produce acid and turn the medium yellow and turbid after incubation at 45°C are presumptive enterococci.

Enterococcosel™ Agar

Composition per liter:

Pancreatic digest of casein	17.0g
Agar	13.5g
Oxgall	10.0g
NaCl	5.0g
Yeast extract	5.0g
Peptic digest of animal tissue	3.0g
Esculin	1.0g
Sodium citrate	1.0g
Ferric ammonium citrate	0.5g
NaN ₃	0.25g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the rapid, selective isolation, cultivation, and enumeration of fecal group D streptococci (enterococci). For the cultivation of staphylococci and *Listeria monocytogenes*.

Enterococcosel Broth

Composition per liter:

Pancreatic digest of casein	17.0g
Oxgall	10.0g
NaCl	5.0g
Yeast extract	5.0g
Peptic digest of animal tissue	3.0g
Esculin	1.0g
Sodium citrate	1.0g
Ferric ammonium citrate	0.5g
NaN ₃	0.25g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until dissolved. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of group D streptococci (enterococci).

Enterococcus Agar (m-Enterococcus Agar) (Azide Agar)

Composition per liter:

Pancreatic digest of casein	15.0g
Agar	10.0g
Papaic digest of soybean meal	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	4.0g
Glucose	2.0g
NaN ₃	0.4g
Triphenyltetrazolium chloride	0.1g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 45°–50°C. Do not autoclave. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and enumeration of enterococci in water, sewage and feces by the membrane filter method. For the direct plating of specimens for the detection and enumeration of fecal streptococci.

Esculin Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	13.0g
NaCl	5.0g
Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
Esculin	1.0g
Ferric citrate	0.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 3.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of bacteria based on their ability to hydrolyze esculin and produce H₂S. Bacteria that hydrolyze esculin appear as colonies surrounded by a reddish-brown to dark brown zone. Bacteria that produce H₂S appear as black colonies.

Esculin Agar, Modified CDC (BAM M53)

Composition per liter:

Heart muscle, infusion from	375.0g
Agar	15.0g
Thiotone	10.0g
NaCl	5.0g
Esculin	1.0g
Ferric citrate	0.5g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 55°C. Adjust pH to 7.0. Distribute into tubes

or leave in flask. Autoclave for 20 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in inclined position to produce slants.

Use: For the differentiation of *Enterobacter* spp.

Esculin Broth

Composition per liter:

Beef heart, solids from infusion	500.0g
Tryptose	10.0g
NaCl	5.0g
Agar	1.0g
Esculin	1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 7.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to hydrolyze esculin. Bacteria that hydrolyze esculin turn the medium brown-black to black.

Esculin Iron Agar

Composition per liter:

Agar	15.0g
Esculin	1.0g
Ferric ammonium citrate	0.5g

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and identification of enterococci based on their ability to hydrolyze esculin. Used in conjunction with E agar and the membrane filter method.

Esculin Mannitol Agar

Composition per liter:

Agar	13.5g
Polypeptone	10.0g
D-Mannitol	10.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g

NaCl.....	5.0g
Heart peptone.....	3.0g
Cornstarch.....	1.0g
Esculin.....	1.0g
Ferric ammonium citrate.....	0.5g
Phenol Red.....	0.025g
Nalidixic acid solution.....	10.0mL
Colistin solution.....	10.0mL
pH 7.3 ± 0.2 at 25°C	

Nalidixic Acid Solution:**Composition per 10.0mL:**

Nalidixic acid.....	0.015g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Colistin Solution:**Composition per 10.0mL:**

Colistin.....	0.01g

Preparation of Colistin Solution: Add colistin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except nalidixic acid solution and colistin solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile nalidixic acid solution and colistin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of *Staphylococcus aureus* and group D streptococci based on mannitol fermentation and hydrolysis of esculin. Bacteria that ferment mannitol appear as yellow colonies surrounded by a yellow zone. Bacteria that hydrolyze esculin appear as dark brown to black colonies surrounded by a dark brown to black zone.

ETGPA**(Egg Tellurite Glycine Pyruvate Agar)****Composition per liter:**

Agar.....	17.0g
Glycine.....	12.0g
Sodium pyruvate.....	10.0g
Pancreatic digest of casein.....	10.0g
Beef extract.....	5.0g
LiCl.....	5.0g
Yeast extract.....	1.0g

Egg yolk emulsion.....	50.0mL
K ₂ TeO ₃ solution.....	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Egg Yolk Emulsion:**Composition:**

Chicken egg yolks.....	11
Whole chicken egg.....	1

Preparation of Egg Yolk Emulsion: Soak egg with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

K₂TeO₃ Solution:**Composition per 100.0mL:**

K ₂ TeO ₃	1.0g

Preparation of K₂TeO₃ Solution: Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Add 10.0mL of sterile 1% tellurite solution and 50.0mL of sterile egg yolk emulsion. If desired, add sulfamethazine to a final concentration of 50.0mg/mL. Mix thoroughly but gently and pour into sterile Petri dishes.

Use: For the selective isolation and enumeration of coagulase-positive staphylococci from food. For the differentiation and identification of staphylococci on the basis of their ability to clear egg yolk. Addition of sulfamethazine inhibits the growth of *Proteus*. Gray-black colonies surrounded by a clear zone are diagnostic for *Staphylococcus aureus*.

**Ethyl Violet Azide Broth
(EVA Broth)****Composition per liter:**

Pancreatic digest of casein.....	13.5g
Yeast extract.....	6.5g
Glucose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.7g
KH ₂ PO ₄	2.7g
NaN ₃	0.4g
Ethyl Violet.....	0.83mg
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Use: For the isolation, cultivation, and enumeration of enterococci. Fecal enterococci turn the medium turbid with a purple sediment on the bottom of the tube.

Ethyl Violet Azide Broth (EVA Broth)

Composition per liter:

Tryptose	20.0g
Glucose	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.7g
KH ₂ PO ₄	2.7g
NaN ₃	0.4g
Ethyl Violet.....	0.83mg
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Use: For the isolation, cultivation, and enumeration of enterococci. Fecal enterococci turn the medium turbid with a purple sediment on the bottom of the tube.

Ethyl Violet Azide Broth (EVA Broth)

Composition per liter:

Tryptose	20.0g
Glucose	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.7g
KH ₂ PO ₄	2.7g

NaN ₃	0.3g
Ethyl Violet.....	0.5mg
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Use: For the isolation, cultivation, and enumeration of enterococci. Fecal enterococci turn the medium turbid with a purple sediment on the bottom of the tube.

Fay and Barry Medium

Composition per liter:

Amino acid.....	10.0g
Peptone	5.0g
Yeast extract.....	3.0g
Bromcresol Purple solution	5.0mL
pH 5.5 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.2g
Ethanol.....	50.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to 50.0mL of absolute ethanol. Add distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. The amino acid may be L-arginine, L-ornithine, or L-lysine, depending on which amino acid decarboxylase activity is being measured. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the determination of decarboxylase activities of *Aeromonas* species.

FC Agar (Fecal Coliform Agar) (m-FC Agar) (m-Fecal Coliform Agar)

Composition per liter:

Agar	15.0g
Lactose.....	12.5g
NaCl.....	5.0g

Proteose peptone No. 3	5.0g
Yeast extract	3.0g
Bile salts	1.5g
Aniline Blue	0.1g
Rosolic acid solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Rosolic Acid Solution:

Composition per 100.0mL:

Rosolic acid	1.0g

Preparation of Rosolic Acid Solution: Add rosolic acid to 0.2N NaOH and bring volume to 100.0L. Mix thoroughly.

Preparation of Medium: Add 10.0mL rosolic acid solution to 950.0mL distilled/deionized water. Mix thoroughly. Add other components and bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling with frequent mixing. Do not autoclave. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of coliform bacteria using the membrane filtration method.

FC Agar
(Fecal Coliform Agar)
(m-FC Agar)
(m-Fecal Coliform Agar)

Composition per liter:

Agar	15.0g
Lactose	12.5g
Tryptose	10.0g
NaCl	5.0g
Proteose peptone No. 3	5.0g
Yeast extract	3.0g
Bile salts	1.5g
Aniline Blue	0.1g
Rosolic acid solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Rosolic Acid Solution:

Composition per 100.0mL:

Rosolic acid	1.0g

Preparation of Rosolic Acid Solution: Add rosolic acid to 0.2N NaOH and bring volume to 100.0L. Mix thoroughly.

Preparation of Medium: Add 10.0mL rosolic acid solution to 950.0mL of distilled/deionized water. Mix thoroughly. Add other components and bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling with fre-

quent mixing. Do not autoclave. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of coliform bacteria using the membrane filtration method.

FC Broth
(Fecal Coliform Broth)
(m-FC Broth)
(m-Fecal Coliform Broth)

Composition per liter:

Lactose	12.5g
Tryptose	10.0g
NaCl	5.0g
Proteose peptone No. 3	5.0g
Yeast extract	3.0g
Bile salts	1.5g
Aniline Blue	0.1g
Rosolic acid solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Rosolic Acid Solution:

Composition per 100.0mL:

Rosolic acid	1.0g

Preparation of Rosolic Acid Solution: Add rosolic acid to 0.2N NaOH and bring volume to 100.0L. Mix thoroughly.

Preparation of Medium: Add 10.0mL of rosolic acid solution to 950.0mL of distilled/deionized water. Mix thoroughly. Add other components and bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling with frequent mixing. Do not autoclave. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of coliform bacteria using the membrane filtration method.

Fecal Coliform Agar, Modified
(m-Fecal Coliform Agar, Modified)
(FCIC)

Composition per liter:

Agar	15.0g
Inositol	10.0g
Tryptose	10.0g
Proteose peptone No. 3	5.0g
NaCl	5.0g
Yeast extract	3.0g
Bile salts No. 3	1.5g
Aniline Blue	0.1g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 50°C.

Adjust pH to 7.4. Pour into sterile Petri dishes in 20.0mL volumes. Allow surface of plates to dry before using.

Use: For the isolation, cultivation, and enumeration of *Klebsiella* species using the membrane filter method.

Fecal Coliform Agar, Modified

Composition per liter:

Agar	15.0g
Lactose	12.5g
Tryptose	10.0g
Proteose peptone No. 3	5.0g
NaCl	5.0g
Yeast extract	3.0g
Bile salts No. 3	1.5g
Aniline Blue	0.1g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 50°C. Adjust pH to 7.4. Pour into sterile Petri dishes in 20.0mL volumes. Allow surface of plates to dry before using.

Use: For the isolation, cultivation, and identification of stressed fecal coliform microorganisms based on their ability to ferment lactose. Lactose-fermenting bacteria turn the medium blue.

Fermentation Basal Medium

Composition per liter:

Agar	15.0g
(NH ₄) ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.2g
KCl	0.02g
Carbohydrate solution	100.0mL
Bromcresol Purple solution	20.0mL

pH 7.0 ± 0.2 at 25°C

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	0.04g
Ethanol	50.0mL

Preparation of Bromcresol Purple Solution: Add Bromcresol Purple to 50.0mL of absolute ethanol. Add distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Various carbohydrates are used for different fermentation tests. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the differentiation of aerobic actinomycetes based upon carbohydrate fermentation. Actinomycetes that produce acid from carbohydrates turn the medium yellow.

Fermentation Broth (CHO Medium)

Composition per liter:

Pancreatic digest of casein	15.0g
Yeast extract	7.0g
NaCl	2.5g
Agar	0.75g
Sodium thioglycolate	0.5g
L-Cystine	0.25g
Ascorbic acid	0.1g
Bromthymol Blue	0.01g
Carbohydrate or starch solution	100.0mL

pH 7.0 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate	6.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Starch Solution:

Composition per 100.0mL:

Starch	2.5g

Preparation of Starch Solution: Add starch to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks. Loosen caps on tubes. Place in an anaerobic chamber under an atmosphere of 85% N₂, 10% H₂, and 5% CO₂. Fasten the caps securely or maintain in an anaerobic chamber.

Use: For the differentiation of anaerobic bacteria based upon carbohydrate fermentation. Bacteria that ferment carbohydrates turn the medium yellow.

Fermentation Medium

Composition per liter:

Glucose or mannitol.....	10.0g
Pancreatic digest of casein.....	10.0g
Agar	2.2g
Yeast extract.....	1.0g
Bromocresol Purple	0.04g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For differentiating *Staphylococcus* and *Micrococcus* species based upon the fermentation of glucose and mannitol.

FGTC Agar

Composition per liter:

Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g
NaCl.....	5.0g
KH ₂ PO ₄	5.0g
Amylose Azure	3.0g
Galactose.....	1.0g
Thallous acetate	0.5g
MUG (4-Methylumbelliferyl- α -D-galactoside)...	0.1g
NaHCO ₃ solution	20.0mL
Gentamicin solution.....	2.5mL
Tween 80.....	0.75mL
pH 7.3 ± 0.2 at 25°C	

Gentamicin Solution:

Composition per 10.0mL:

Gentamicin.....	0.01g

Preparation of Gentamicin Solution: Add gentamicin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

NaHCO₃ Solution:

Composition per 20.0mL:

NaHCO ₃	2.0g

Preparation of NaHCO₃ Solution: Add the NaHCO₃ to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: Add components, except NaHCO₃ solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly.

Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add sterile NaHCO₃ solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation, differentiation, and enumeration of *Enterococcus* species based on starch hydrolysis and production of fluorescence. Bacteria that hydrolyze starch, such as *Streptococcus bovis*, appear as colonies surrounded by a clear zone. Bacteria that produce fluorescence, such as *Streptococcus bovis* and *Enterococcus faecium*, appear as colonies surrounded by a zone of bright bluish fluorescence when viewed under a long-wave UV lamp. Other bacteria, such as *Enterococcus faecalis*, *Enterococcus avium*, or *Streptococcus equinus*, do not hydrolyze starch or produce fluorescence.

Fish Peptone Agar

Composition per liter:

Agar	5.0g
Maltose	5.0g
NaCl.....	5.0g
Peptone	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	5.0g
Trout tissue extract solution.....	50.0mL
pH 7.0 ± 0.2 at 25°C	

Trout Tissue Extract Solution:

Composition per liter:

Fish (brook trout).....	500.0g
Pepsin.....	1.0g
HCl, concentrated	15.0mL

Preparation of Trout Tissue Extract: Add 1.0L of distilled/deionized water to brook trout and blend for 20–30 min. Add 1.0g of pepsin and 15.0mL of concentrated HCl to digest the trout proteins. Incubate for 12 hr at 45°C. Adjust pH to 7.0. Allow solids to settle. Filter sterilize. Do not autoclave. Store at 5°C.

Preparation of Medium: Add components, except trout tissue extract, to distilled/deionized water and bring volume to 950.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile trout tissue extract. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Aeromonas salmonicida*.

Fish Peptone Broth

Composition per liter:

Maltose	5.0g
NaCl.....	5.0g
Peptone	5.0g

Pancreatic digest of casein.....	5.0g
Yeast extract.....	5.0g
Trout tissue extract solution.....	50.0mL
pH 7.0 ± 0.2 at 25°C	

Trout Tissue Extract Solution:

Composition per liter:

Fish (brook trout).....	500.0g
Pepsin.....	1.0g
HCl, concentrated.....	15.0mL

Preparation of Trout Tissue Extract: Add 1.0L of distilled/deionized water to brook trout and blend for 20–30 min. Add 1.0g of pepsin and 15.0mL of concentrated HCl to digest the trout proteins. Incubate for 12 hr at 45°C. Adjust pH to 7.0. Allow solids to settle. Filter sterilize. Do not autoclave. Store at 5°C.

Preparation of Medium: Add components, except trout tissue extract, to distilled/deionized water and bring volume to 950.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 10 psi pressure–118°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile trout tissue extract. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Aeromonas salmonicida*.

Flavobacterium Medium (ATCC Medium 647)

Composition per liter:

Agar.....	12.0g
Beef extract.....	10.0g
Peptone.....	10.0g
NaCl.....	5.0g
pH 7.2–7.3 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Flavobacterium* species from food and food-processing equipment.

Flavobacterium Medium (ATCC Medium 1687)

Composition per liter:

Sodium glutamate.....	4.0g
K ₂ HPO ₄	0.65g
NaNO ₃	0.5g
KH ₂ PO ₄	0.19g
MgSO ₄ ·7H ₂ O.....	0.1g
FeSO ₄ solution.....	2.0mL
pH 7.4 ± 0.2 at 25°C	

FeSO₄ Solution:

Composition per 10.0mL:

FeSO ₄ ·7H ₂ O.....	0.03g

Preparation of FeSO₄ Solution: Add FeSO₄ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except FeSO₄ solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 2.0mL of sterile FeSO₄ solution. Mix thoroughly. Adjust pH to 7.4. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Flavobacterium* species.

Fluid Sabouraud Medium

Composition per liter:

Glucose.....	20.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
pH 5.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For cultivation of yeasts, molds, and aciduric microorganisms.

Fluid Thioglycolate Agar

Composition per liter:

Pancreatic digest of casein.....	15.0g
Glucose.....	5.0g
Yeast extract.....	5.0g
NaCl.....	2.5g
Agar.....	0.75g
L-Cystine.....	0.5g
Sodium thioglycolate.....	0.5g
Resazurin.....	1.0mg
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of anaerobic bacteria. For the cultivation of *Campylobacter fetus*, *Campylobacter jejuni*, and *Streptococcus* species.

Fluid Thioglycolate Agar with Calcium Carbonate

Composition per liter:

Agar	75.0g
Pancreatic digest of casein	15.0g
CaCO ₃	10.0g
Glucose	5.0g
Yeast extract	5.0g
NaCl	2.5g
L-Cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin	0.001g

pH 7.1 ± 0.2 at 25°C

Preparation of Medium: Add components, except CaCO₃, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Sterilize CaCO₃ by autoclaving for 15 min at 15 psi pressure–121°C. Aseptically add sterile CaCO₃ to sterile tubes or plates—0.1g of sterile CaCO₃ per 10.0mL of medium to be added. Pour medium into sterile Petri dishes or distribute into sterile tubes.

Use: For the rapid cultivation of anaerobic bacteria. For the cultivation and maintenance of *Campylobacter fetus*, *Campylobacter jejuni*, and *Streptococcus* species.

Fletcher Medium

Composition per liter:

Agar	1.5g
NaCl	0.5g
Peptone	0.3g
Beef extract	0.2g
Rabbit serum	50.0mL

pH 7.9 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except rabbit serum, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 50.0mL of sterile rabbit serum. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation, cultivation, and maintenance of cultures of *Leptospira* species.

Fletcher Medium with Fluorouracil (Fluorouracil *Leptospira* Medium)

Composition per liter:

Agar	1.5g

NaCl	0.5g
Peptone	0.3g
Beef extract	0.2g
Rabbit serum	50.0mL
Fluorouracil solution	20.0mL
pH 7.9 ± 0.1 at 25°C	

Fluorouracil Solution:

Composition per 100.0mL:

Fluorouracil	10.0g

Preparation of Fluorouracil Solution: Add fluorouracil to 50.0mL of distilled/deionized water. Add 1.0mL of 2N NaOH and bring volume to 100.0mL. Gently heat to 56°C for 2 hr. Adjust pH to 7.4–7.6 with NaOH. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except rabbit serum and fluorouracil solution, to distilled/deionized water and bring volume to 930.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 80.0mL of sterile rabbit serum. Mix thoroughly. Aseptically distribute into sterile tubes or flasks. Immediately prior to use, add 0.1mL of fluorouracil solution per 5.0mL of medium.

Use: For the isolation, cultivation, and maintenance of cultures of *Leptospira* species.

Fletcher's Semisolid Medium

Composition per 2120.0mL:

Agar	1.5g
NaCl	0.5g
Peptone	0.3g
Beef extract	0.2g
Rabbit serum	240.0mL

pH 7.9 ± 0.1 at 25°C

Preparation of Medium: Add components, except rabbit serum, to distilled/deionized water and bring volume to 1880.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 240.0mL of sterile rabbit serum. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation, cultivation, and maintenance of cultures of *Leptospira* species.

FLN Medium

(Fluorescence Lactose Nitrate Medium)

Composition per liter:

Lactose	20.0g
Agar	15.0g
Proteose peptone No. 3	10.0g
KNO ₃	2.0g
K ₂ HPO ₄	1.5g

MgSO ₄ ·7H ₂ O	1.5g
NaNO ₂	0.5g
Phenol Red	0.02g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of pseudomonads from other nonfermentative bacilli. Lactose fermentation is indicated by the medium turning yellow. *Pseudomonas cepacia* often produces acid from lactose. Denitrification from nitrate or nitrite is indicated by the formation of gas bubbles in the solid medium. *Pseudomonas aeruginosa*, *Pseudomonas mendocina*, and *Pseudomonas denitrificans* are positive for denitrification. Fluorescein production is indicated by fluorescence under UV light. *Pseudomonas aeruginosa* is positive for fluorescein production; *Pseudomonas denitrificans* does not produce fluorescein.

Flo Agar

Composition per liter:

Agar	14.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	1.5g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For cultivation of fluorescent *Pseudomonas* species.

Fluid Thioglycolate Agar with Calcium Carbonate

Composition per liter:

Agar	75.0g
Pancreatic digest of casein	15.0g
CaCO ₃	10.0g
Glucose	5.0g
Yeast extract	5.0g
NaCl	2.5g
L-Cystine	0.5g

Sodium thioglycolate	0.5g
Resazurin	0.001g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components, except CaCO₃, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Sterilize CaCO₃ by autoclaving for 15 min at 15 psi pressure–121°C. Aseptically add sterile CaCO₃ to sterile tubes or plates—0.1g of sterile CaCO₃ per 10.0mL of medium to be added. Pour medium into sterile Petri dishes or distribute into sterile tubes.

Use: For the rapid cultivation of anaerobic bacteria. For the cultivation and maintenance of *Campylobacter fetus*, *Campylobacter jejuni*, *Leptotrichia buccalis*, and *Streptococcus* species.

Fluid Thioglycolate Medium

Composition per liter:

Pancreatic digest of casein	15.0g
Glucose	5.5g
Yeast extract	5.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin	1.0mg
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use (resazurin turns red), heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of anaerobic, microaerophilic, and aerobic microorganisms. For use in sterility testing of a variety of specimens.

Fluid Thioglycolate Medium with Beef Extract

Composition per liter:

Pancreatic digest of casein	15.0g
Glucose	5.5g
Yeast extract	5.0g
Beef extract	5.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.5g

Sodium thioglycolate	0.5g
Resazurin	1.0mg
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use (resazurin turns red), heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of anaerobic, microaerophilic, and aerobic microorganisms. For use in sterility testing of a variety of specimens.

Fluid Thioglycolate Medium with K Agar

Composition per liter:

Pancreatic digest of casein	15.0g
Glucose	5.0g
Yeast extract	5.0g
KCl	2.5g
K agar	0.45g
L-Cystine	0.5g
Resazurin	1.0mg
Thioglycolic acid	0.3mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use (resazurin turns red), heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of various microorganisms. For use in sterility test.

Fluid Thioglycolate Medium with Rabbit Serum

Composition per liter:

Pancreatic digest of casein	15.0g
Glucose	5.5g
Yeast extract	5.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin	1.0mg
Rabbit serum	100.0mL
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except rabbit serum, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 100.0mL rabbit serum. Aseptically distribute into sterile screw-capped tubes or flasks.

Use: For cultivation of anaerobic bacteria. For use in sterility testing.

Fluid Thioglycolate Medium without Glucose or E_h Indicator

Composition per liter:

Pancreatic digest of casein	20.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.5g
Sodium thioglycolate	0.5g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically distribute into sterile screw-capped tubes or flasks.

Use: For cultivation of anaerobic bacteria. For use as a basal medium in carbohydrate fermentation tests for differentiating anaerobic bacteria. For carbohydrate fermentation tests, 1.0mL of a 10% filter-sterilized carbohydrate solution is aseptically added to 9.0mL of fluid thioglycolate medium.

Fluorescent Pectolytic Agar (FPA Medium)

Composition per liter:

Proteose peptone No. 3	20.0g
Agar	15.0g
Pectin	5.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	0.73g
Antibiotic solution	10.0mL

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide	0.075g
Novobiocin	0.045g
Penicillin G	75,000U
Ethanol	1.0mL

Preparation of Antibiotic Solution: Add components to 1.0mL of ethanol. Mix thoroughly. Let stand for 30 min. Bring volume to 10.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of fluorescent *Pseudomonas* species that are pectinolytic.

Fluorocult *E. coli* O157:H7 Agar (*E. coli* O157:H7 Agar, Fluorocult)

Composition per liter:

Peptone from casein.....	20.0g
Agar	13.0g
Sorbitol.....	10.0g
NaCl.....	5.0g
Meat extract	2.0g
Na ₂ S ₂ O ₃	2.0g
Sodium deoxycholate.....	1.12g
Yeast extract.....	1.0g
Ammonium ferric citrate	0.5g
4-Methylumbelliferyl-β-D-glucuronide	0.1g
Bromthymol Blue	0.025g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the isolation and differentiation of enterohemorrhagic (EHEC) *Escherichia coli* O157:H7 strains from foods. In contrast to most other *E. coli* strains, *E. coli* O157:H7 shows the following characteristics: no sorbitol-cleavage capacity within 48 hr. and no formation of glucuronidase (MUG-negative/no fluorescence). Sodium deoxycholate inhibits the growth of the Gram-positive accompanying flora for the greater part. Sorbitol serves, together with the pH indicator bromthymol blue, to determine the degradation of sorbitol which, in the case of sorbitol-positive microorganisms, results in the colonies turning yellow in color. Sorbitol-negative strains, on the other hand, do not lead to any change in the color of the culture medium and thus proliferate as greenish colonies. Sodium thiosulfate and ammonium iron(III) citrate result in black-brown discoloration of the agar for colonies, in the presence of hydrogen-sulfide-forming pathogens, precipitating iron sulfide. *Pro-*

teus mirabilis in particular, which displays biochemical properties similar to those of *E. coli* O157:H7, can thus be very easily differentiated from *E. coli* O157:H7 on account of the brownish discoloration. 4-methylumbelliferyl-β-D-glucuronide (MUG) is converted into 4-methylumbelliferone by β-D-glucuronidase-forming pathogens; 4-methylumbelliferone fluoresces under UV light. The activity of β-D-glucuronidase is a highly specific characteristic of *E. coli*. In contrast to most *E. coli* strains, *E. coli* O157:H7 is not capable of forming β-D-glucuronidase. When irradiated with long-wave UV light, no fluorescence is formed.

FN Medium

(Fluorescence Denitrification Medium)

Composition per liter:

Agar	15.0g
Proteose peptone No. 3	10.0g
KNO ₃	2.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	1.5g
NaNO ₂	0.5g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of pseudomonads from other nonfermentative bacilli. Denitrification from nitrate or nitrite is indicated by the formation of gas bubbles in the solid medium. Fluorescein production is indicated by fluorescence under UV light.

Folic Acid Agar

Composition per liter:

Noble agar.....	15.0g
K ₂ HPO ₄ ·3H ₂ O	1.2g
Folic acid	1.0g
KH ₂ PO ₄	0.5g
Salts A.....	5.0mL
Salts B.....	1.5mL

Salts A:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O	1.0g
CaCl ₂ ·2H ₂ O	0.1g
FeSO ₄ ·7H ₂ O.....	0.1g

Preparation of Salts A Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Maintain for 3 days at 25°C to dissolve. Filter sterilize the supernatant.

Salts B:**Composition** per 100.0mL:

MnSO ₄	0.1g
Na ₂ MoO ₄	0.1g

Preparation of Salts B Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except salts A solution and salts B solution, to distilled/deionized water and bring volume to 994.5mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.0mL of sterile salts A solution and 1.5mL of sterile salts B solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Pseudomonas* species.

Folic Acid Assay Medium**Composition** per liter:

Glucose	40.0g
Sodium citrate	20.0g
Vitamin assay casamino acids	12.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
L-Cystine	0.2g
DL-Tryptophan	0.2g
Adenine sulfate	0.02g
FeSO ₄ ·7H ₂ O	0.02g
Guanine-HCl	0.02g
MnSO ₄ ·7H ₂ O	0.02g
NaCl	0.02g
Uracil	0.02g
Pyridoxine-HCl	4.0mg
Niacin	2.0mg
Riboflavin	2.0mg
Thiamine-HCl	2.0mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Biotin	0.8µg

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling with frequent mixing. Cool to 45°–50°C. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the microbiological assaying of folic acid using *Enterococcus hirae* as the test organism.

FPA Medium**Composition** per liter:

Proteose peptone No. 3	20.0g
Agar	15.0g
Pectin, citrus	5.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	1.5g
Antibiotic solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Antibiotic Solution:**Composition** per 10.0mL:

Cycloheximide	0.075g
Novobiocin	0.045g
Penicillin G	75,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except agar and antibiotic solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 7.0. Add agar. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Pseudomonas* species.

Fraser Broth**Composition** per liter:

NaCl	20.0g
Na ₂ HPO ₄	12.0g
Beef Extract	5.0g
Proteose peptone	5.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
LiCl	3.0g
KH ₂ PO ₄	1.35g
Esculin	1.0g
Fraser supplement solution	10.0mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Fraser Supplement Solution:**Composition** per 10.0mL:

Ferric ammonium citrate	0.5g
Acriflavine-HCl	0.25g
Nalidixic acid	0.1g
Ethanol	5.0mL

Preparation of Fraser Supplement Solution: Add components to distilled/deionized

water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except Fraser supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile Fraser supplement solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation of *Listeria* species from foods.

Fraser Secondary Enrichment Broth

Composition per liter:

NaCl.....	20.0g
Na ₂ HPO ₄	12.0g
Beef extract.....	5.0g
Proteose peptone.....	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	5.0g
LiCl.....	3.0g
KH ₂ PO ₄	1.35g
Esculin.....	1.0g
Acriflavin solution.....	10.0mL
Ferric ammonium citrate solution.....	10.0mL
Nalidixic acid solution.....	1.0mL

Ferric Ammonium Citrate Solution:

Composition per 10.0mL:

Ferric ammonium citrate.....	0.5g

Preparation of Ferric Ammonium Citrate Solution: Add ferric ammonium citrate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Acriflavin Solution:

Composition per 10.0mL:

Acriflavin.....	0.025g

Preparation of Acriflavin Solution: Add acriflavin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid.....	0.04g
NaOH (0.1N solution).....	10.0mL

Preparation of Nalidixic Acid Solution: Add nalidixic acid to 10.0mL of NaOH solution. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except acriflavin solution and ferric ammonium citrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL vol-

umes. Autoclave for 12 min at 15 psi pressure–121°C. Cool rapidly to 25°C. Immediately prior to inoculation, aseptically add 0.1mL of sterile acriflavin solution and 0.1mL of ferric ammonium citrate solution to each tube. Mix thoroughly.

Use: For the isolation, cultivation, and enrichment of *Listeria monocytogenes* from foods based on esculin hydrolysis. Bacteria that hydrolyze esculin appear as black colonies.

Freezing Medium (BAM M30b)

Composition per 11.5mL:

Bolton broth base.....	9.5mL
Fetal bovine serum.....	1.0mL
Glycerol solution.....	1.0mL

Bolton Broth Base:

Composition per liter:

Peptone.....	10.0g
Lactalbumin hydrolysate.....	5.0g
Yeast extract.....	5.0g
NaCl.....	5.0g
a-ketoglutarate.....	1.0g
Na-pyruvate.....	0.5g
Na-metabisulfite.....	0.5g
Na ₂ CO ₃	0.6g
Hemin.....	0.01g

Preparation of Bolton Broth Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature.

Glycerol Solution:

Composition per 100.0mL:

Glycerol.....	10.0g

Preparation of Glycerol Solution: Add glycerol to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature.

Preparation of Medium: Aseptically combine 9.5mL Bolton broth base, 1.0mL sterile glycerol solution, and 1.0mL filter sterilized fetal bovine serum. Mix thoroughly.

Use: For the preservation of *Campylobacter* spp.

G Medium

Composition per liter:

(NH ₄) ₂ SO ₄	2.0g
Yeast extract.....	2.0g
Glucose.....	1.0g
K ₂ HPO ₄	0.6g
KH ₂ PO ₄	0.4g
MgSO ₄ ·7H ₂ O.....	0.2g

CaCl ₂	0.08g
MnSO ₄ ·H ₂ O.....	0.05g
CuSO ₄ ·5H ₂ O.....	5.0mg
ZnSO ₄ ·7H ₂ O.....	5.0mg
FeSO ₄ ·7H ₂ O.....	0.5mg

pH 7.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Bacillus cereus*.

GBNA Medium

(Gum Base Nalidixic Acid Medium)

Composition per liter:

Gellan gum.....	8.0g
Pancreatic digest of casein.....	5.7g
NaCl.....	1.7g
Papaic digest of soybean meal.....	1.0g
Glucose.....	0.83g
K ₂ HPO ₄	0.83g
MgCl ₂ ·6H ₂ O.....	0.33g
Nalidixic acid.....	0.05g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes*.

Gelatin Agar

Composition per liter:

Gelatin.....	30.0g
Agar.....	15.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	10.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of bacteria isolated from foods and their differentiation based on proteolytic activity.

Gelatin Agar (BAM M54)

Composition per liter:

Agar.....	15.0g
Gelatin.....	15.0g
Peptone.....	4.0g
Yeast extract.....	1.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of heterotrophic bacteria based upon their utilization of gelatin.

Gelatin Infusion Broth

Composition per liter:

Beef heart, solids from infusion.....	500.0g
Gelatin.....	40.0g
Tryptose.....	10.0g
NaCl.....	5.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and differentiation of a variety of heterotrophic bacteria based upon their production of gelatinase. The gelatinase liquifies the medium.

Gelatin Medium

Composition per liter:

Gelatin.....	120.0g
Pancreatic digest of casein.....	13.0g
Sodium chloride.....	5.0g
Yeast extract.....	5.0g
Heart muscle, solids from infusion.....	2.0g
Sodium thioglycolate.....	0.5g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of gelatin-utilizing *Clostridium* species.

Gelatin Phosphate Salt Agar (GPS Agar)

Composition per liter:

Agar	15.0g
Gelatin.....	10.0g
NaCl.....	10.0g
K ₂ HPO ₄	5.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and differentiation of *Vibrio* species from foods.

Gelatin Phosphate Salt Broth (GPS Broth)

Composition per liter:

Gelatin.....	10.0g
NaCl.....	10.0g
K ₂ HPO ₄	5.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio* species from foods.

Gelatin Salt Agar (GS Agar) (BAM M55)

Composition per liter:

NaCl.....	30.0g
Agar	15.0g
Gelatin.....	15.0g
Peptone.....	4.0g
Yeast extract.....	1.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and differentiation of *Vibrio* species from foods.

Gelatin Salt Agar (GS Agar) (BAM M55)

Composition per liter:

NaCl.....	30.0g
Agar	25.0g
Gelatin.....	15.0g
Peptone	4.0g
Yeast extract.....	1.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and differentiation of *Vibrio* species from foods. The high concentration of agar inhibits the spreading of *V. alginolyticus* and some other *Vibrio* spp.

Gelatinase Test Medium

Composition per liter:

Gelatin.....	3.0g
ACES buffer	1.0g
Yeast extract.....	1.0g
Charcoal, activated	0.15g
α-Ketoglutarate monopotassium salt.....	0.1g
L-Cysteine-HCl·H ₂ O (4% solution).....	1.0mL
KOH (85% solution).....	1.0mL
Fe ₄ (P ₂ O ₇) ₃ solution.....	1.0mL

pH 6.9 ± 0.2 at 25°C

L-Cysteine-HCl·H₂O Solution:

Composition per 10.0mL:

L-Cysteine-HCl·H ₂ O.....	0.4g

Preparation of L-Cysteine-HCl·H₂O Solution:

Add L-cysteine-HCl·H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Fe₄(P₂O₇)₃ Solution:

Composition per 10.0mL:

Fe ₄ (P ₂ O ₇) ₃	0.15g

Preparation of Fe₄(P₂O₇)₃ Solution: Add Fe₄(P₂O₇)₃ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add ACES buffer to distilled/deionized water and bring volume to 899.0mL. Mix thoroughly. Gently heat to 50°C. Add 1.0mL of KOH solution. Mix thoroughly. Add charcoal, yeast extract, and α-ketoglutarate. Add 80.0mL of distilled/deionized water to wash sides of flask. Mix thoroughly. Autoclave for 15 min at 15 psi pres-

sure–121°C. Cool to 50°C. Aseptically add 10.0mL of sterile cysteine solution and 10.0mL of sterile $\text{Fe}_4(\text{P}_2\text{O}_7)_3$ solution. Mix thoroughly. Adjust pH to 6.9. Aseptically distribute into sterile screw-capped tubes.

Use: For the cultivation and differentiation of gelatinase-producing bacteria.

Gentamicin Sulfate Solution (BAM M57)

Composition per 100mL:

Gentamicin sulfate 0.5g

Preparation of Gentamicin Sulfate Solution: Add gentamicin sulfate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Use: As an inhibitor to render media selective.

Giolitti-Cantoni Broth (Giolitti-Cantoni Broth Base with Tellurite)

Composition per 1030.0mL:

Mannitol 20.0g
Pancreatic digest of casein 10.0g
Beef extract 5.0g
LiCl 5.0g
NaCl 5.0g
Yeast extract 5.0g
Sodium pyruvate 3.0g
Glycine 1.2g
Chapman tellurite solution 0.3mL

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid and BD Diagnostic Systems.

Chapman Tellurite Solution:

Composition per 100.0mL:

K_2TeO_3 3.5g

Preparation of Chapman Tellurite Solution: Add K_2TeO_3 to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components, except Chapman tellurite solution, to distilled/deionized water and bring volume to 1.0L. Gently heat and bring to boiling. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool rapidly to 25°C. Immediately prior to use, incubate tubes at 100°C to expel oxygen. Aseptically add 0.3mL of sterile Chapman tellurite solution to each tube. Mix thoroughly. Cool to 25°C.

Use: For the cultivation and enrichment of *Staphylococcus aureus* from foods.

Gluconate Peptone Broth

Composition per liter:

Potassium gluconate 40.0g
Casein peptone 1.5g
 K_2HPO_4 1.0g
Yeast extract 1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of Gram-negative bacteria based on their ability to oxidize gluconate to 2-ketogluconate. For the differentiation of fluorescent *Pseudomonas* species. After inoculation with bacteria and 48 hr of growth in this medium, Benedict's reagent is added. Bacteria that produce the reducing sugar 2-ketogluconate turn the reagent yellow-orange to orange-red.

Glucose Agar with 25% Glucose

Composition per liter:

Glucose 250.0g
Agar 25.0g
Polypeptone 5.0g
Malt extract 3.0g
Yeast extract 3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of osmophilic yeasts and bacteria.

Glucose Phosphate Broth

Composition per liter:

Peptone 10.0g
 K_2HPO_4 5.0g
Glucose 5.0g

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components, except glucose, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Filter while hot through Whatman filter paper. Cool to 25°C. Adjust pH to 7.5. Add 5.0g of glucose. Mix thoroughly. Distribute into sterile

tubes or flasks. Autoclave for 10 min at 10 psi pressure—115°C.

Use: For the cultivation of a variety of nonfastidious heterotrophic microorganisms.

Glucose Salt Teepol Broth (GSTB)

Composition per liter:

NaCl	30.0g
Peptone	10.0g
Glucose	5.0g
Beef extract	3.0g
Methyl Violet	2.0mg
Sodium lauryl sulfate (Teepol—0.1% solution)	4.0mL
pH 8.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.8. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure—121°C.

Use: For the cultivation of *Vibrio* species from foods.

Glucose Salts Medium

Composition per 1000.5mL:

Glucose	5.0g
(NH ₄) ₂ SO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.5g
NaH ₂ PO ₄ ·12H ₂ O	0.7mg
NaH ₂ PO ₄ ·2H ₂ O	0.3mg
Trace elements solution	0.5mL
pH 6.9 ± 0.2 at 25°C	

Trace Elements Solution:

Composition per liter:

H ₃ BO ₃	0.3g
CoCl ₂ ·6H ₂ O	0.2g
ZnSO ₄ ·7H ₂ O	0.1g
MnCl ₂ ·4H ₂ O	0.03g
Na ₂ MoO ₄ ·2H ₂ O	0.03g
NiCl ₂ ·6H ₂ O	0.02g
CuCl ₂ ·2H ₂ O	0.01g

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except trace elements solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure—121°C. Aseptically add 0.5mL of sterile trace elements solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of a wide variety of bacteria and fungi.

Glucose Yeast Extract Agar

Composition per liter:

Agar	15.0g
Glucose	5.0g
Meat extract	5.0g
Peptone	5.0g
Yeast extract	5.0g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure—121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Leuconostoc* species.

Glucose Yeast Extract Peptone Agar with 2% Glucose

Composition per liter:

Glucose	20.0g
Agar	15.0g
Peptone	5.0g
Yeast extract	1.0g
K ₂ HPO ₄	0.5g
MgSO ₄ ·7H ₂ O	0.5g
CaCO ₃	1.0g
FeSO ₄ ·7H ₂ O	50.0mg
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.8. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure—121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus acidophilus*.

Glucose Yeast Extract Peptone Medium (GYP Medium)

Composition per liter:

Glucose	20.0g
Agar	10.0g
Peptone	5.0g
Yeast extract	5.0g
CaCO ₃	0.1g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Sporolactobacillus* species.

Glucose Yeast Medium with Calcium Carbonate

Composition per liter:

Agar	15.0g
CaCO ₃	7.5g
Peptone	5.0g
Yeast extract	5.0g
Glucose	3.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Adjust pH to 6.3. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Erwinia herbicola* and *Bacillus* species.

Glutamate Medium (ATCC Medium 820)

Composition per liter:

Agar	15.0g
Sodium glutamate	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.2g
KCl	0.1g
Glucose solution	100.0mL

pH 6.5 ± 0.2 at 25°C

Glucose Solution:

Composition per 100.0mL:

Glucose	10.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Pseudomonas* species.

Glutamate Medium (ATCC Medium 1372)

Composition per liter:

K ₂ HPO ₄	6.0g
Sodium glutamate	5.0g
Peptone	4.0g
Yeast extract	4.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

Glutamine Medium

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
Glutamine/tryptophan solution	10.0mL

pH 6.8 ± 0.2 at 25°C

Glutamine/Tryptophan Solution:

Composition per liter:

L-Glutamine	100.0mg
L-Tryptophan	100.0mg

Preparation of Medium: Add components, except glutamine/tryptophan solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile glutamine/tryptophan solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Escherichia coli*.

Glutarate Medium

Composition per liter:

Sodium glutarate	2.6g
NaCl	1.0g
KCl	0.5g
MgCl ₂ ·6H ₂ O	0.4g
NH ₄ Cl	0.25g
KH ₂ PO ₄	0.2g
CaCl ₂ ·2H ₂ O	0.15g
Resazurin	1.0mg
Rumen fluid	20.0mL
NaHCO ₃ solution	20.0mL
Na ₂ S·9H ₂ O solution	10.0mL
Trace elements solution SL-10	1.0mL

pH 7.2 ± 0.2 at 25°C

Trace Elements Solution SL-10:**Composition per liter:**

FeCl ₂ ·4H ₂ O	1.5g
CoCl ₂ ·6H ₂ O	190.0mg
MnCl ₂ ·4H ₂ O	100.0mg
ZnCl ₂	70.0mg
Na ₂ MoO ₄ ·2H ₂ O	36.0mg
NiCl ₂ ·6H ₂ O	24.0mg
H ₃ BO ₃	6.0mg
CuCl ₂ ·2H ₂ O	2.0mg
HCl (25% solution)	10.0mL

Preparation of Trace Elements Solution SL-10:

Add FeCl₂·4H₂O to 10.0mL of HCl solution. Mix thoroughly. Add distilled/deionized water and bring volume to 1.0L. Add remaining components. Mix thoroughly. Gas under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

NaHCO₃ Solution:**Composition per 20.0mL:**

NaHCO ₃	2.5g

Preparation of NaHCO₃ Solution: Add NaHCO₃ to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize. Gas under 80% N₂ + 20% CO₂.

Na₂S·9H₂O Solution:**Composition per 10.0mL:**

Na ₂ S·9H ₂ O	0.36g

Preparation of Na₂S·9H₂O Solution: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Gas under 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Prepare and dispense medium under 80% N₂ + 20% CO₂. Add components, except NaHCO₃ solution and Na₂S·9H₂O solution, to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically and anaerobically add 20.0mL of sterile NaHCO₃ solution and 10.0mL of sterile Na₂S·9H₂O solution. Mix thoroughly. Aseptically and anaerobically distribute into sterile tubes or flasks.

Use: For the cultivation of a wide variety of bacteria that can utilize glutarate as a carbon source.

GN Broth, Hajna**Composition per liter:**

Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Sodium citrate	5.0g
K ₂ HPO ₄	4.0g
D-Mannitol	2.0g

KH ₂ PO ₄	1.5g
Glucose	1.0g
Sodium deoxycholate	0.5g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective cultivation of *Salmonella* and *Shigella* species.

**Green Yeast and Mold Broth
(m-Green Yeast and Mold Broth)****Composition per liter:**

Glucose	50.0g
Yeast extract	9.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
MgSO ₄ ·7H ₂ O	2.1g
K ₂ HPO ₄	2.0g
α-Amylase	0.05g
Thiamine	0.05g
Bromcresol Green	0.026g
pH 4.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the cultivation of fungi from beverages.

GSP Agar**Composition per liter:**

Starch, soluble	20.0g
Agar	15.0g
Sodium glutamate	10.0g
K ₂ HPO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.5g
Phenol Red	0.36g
pH 7.2–7.4 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation and cultivation of *Pseudomonas* species.

Gum Tragacanth Gum Arabic Medium

Composition per 100.0mL:

Gum tragacanth.....	2.0g
Gum arabic.....	1.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation of aciduric flat sour spore-formers from foods. For the isolation and cultivation of *Desulfotomaculum nigrificans* from foods.

GV Medium

Composition per 1061.0mL:

NaHCO ₃	3.0g
NaCl.....	2.25g
Yeast extract.....	1.0g
NH ₄ Cl.....	0.5g
K ₂ HPO ₄	0.348g
KH ₂ PO ₄	0.227g
FeSO ₄ ·7H ₂ O.....	2.0mg
Resazurin.....	0.5mg
Substrate solution.....	20.0mL
Vitamin solution.....	10.0mL
L-Cysteine·HCl solution.....	10.0mL
Na ₂ S·9H ₂ O solution.....	10.0mL
MgSO ₄ ·7H ₂ O solution.....	10.0mL
CaCl ₂ ·2H ₂ O solution.....	10.0mL
Selenite/tungstate solution.....	1.0mL
Trace elements solution SL-10.....	1.0mL
pH 7.0 ± 0.2 at 25°C	

Substrate Solution:

Composition per 20.0mL:

Sodium citrate.....	6.0g

Preparation of Substrate Solution: Add sodium citrate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Vitamin Solution:

Composition per liter:

Pyridoxine·HCl.....	10.0mg
Calcium DL-pantothenate.....	5.0mg
Lipoic acid.....	5.0mg
Nicotinic acid.....	5.0mg
p-Aminobenzoic acid.....	5.0mg
Riboflavin.....	5.0mg
Thiamine·HCl.....	5.0mg
Biotin.....	2.0mg
Folic acid.....	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

L-Cysteine·HCl Solution:

Composition per 10.0mL:

L-Cysteine·HCl.....	0.3g

Preparation of L-Cysteine·HCl Solution: Add L-cysteine·HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Na₂S·9H₂O Solution:

Composition per 10.0mL:

Na ₂ S·9H ₂ O.....	0.3g

Preparation of Na₂S·9H₂O Solution: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Gas under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

MgSO₄·7H₂O Solution:

Composition per 10.0mL:

MgSO ₄ ·7H ₂ O.....	0.5g

Preparation of MgSO₄·7H₂O Solution: Add MgSO₄·7H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

CaCl₂·2H₂O Solution:

Composition per 10.0mL:

CaCl ₂ ·2H ₂ O.....	0.25g

Preparation of CaCl₂·2H₂O Solution: Add CaCl₂·2H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Selenite-Tungstate Solution:

Composition per liter:

NaOH.....	0.5g
Na ₂ WO ₄ ·2H ₂ O.....	4mg
Na ₂ SeO ₃ ·5H ₂ O.....	3mg

Preparation of Selenite-Tungstate Solution:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Trace Elements Solution SL-10:

Composition per liter:

FeCl ₂ ·4H ₂ O.....	1.5g
CoCl ₂ ·6H ₂ O.....	190.0mg
MnCl ₂ ·4H ₂ O.....	100.0mg

ZnCl ₂	70.0mg
Na ₂ MoO ₄ ·2H ₂ O	36.0mg
NiCl ₂ ·6H ₂ O	24.0mg
H ₃ BO ₃	6.0mg
CuCl ₂ ·2H ₂ O	2.0mg
HCl (25% solution).....	10.0mL

Preparation of Trace Elements Solution SL-10:

Add FeCl₂·4H₂O to 10.0mL of HCl solution. Mix thoroughly. Add distilled/deionized water and bring volume to 1.0L. Add remaining components. Mix thoroughly. Gas under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Prepare and dispense medium under 80% N₂ + 20% CO₂. Add components, except substrate solution, MgSO₄·7H₂O solution, CaCl₂·2H₂O solution, selenite/tungstate solution, L-cysteine-HCl solution, and Na₂S·9H₂O solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically and anaerobically add 20.0mL of sterile substrate solution, 10.0mL of sterile MgSO₄·7H₂O solution, 10.0mL of sterile CaCl₂·2H₂O solution, 1.0mL of sterile selenite/tungstate solution, 10.0mL of sterile L-cysteine-HCl solution, and 10.0mL of sterile Na₂S·9H₂O solution. Mix thoroughly. Adjust pH to 7.0 to 7.2. Aseptically and anaerobically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Campylobacter* species.

H Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli* and a variety of other bacteria.

H Broth

Composition per liter:

NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g

Peptone	5.0g
Beef extract.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	1.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 4.0mL volumes. Autoclave for 15 min at 10 psi pressure–115°C.

Use: For the preparation of the H agglutination antigen used in the differentiation and identification of *Salmonella* species types and subtypes.

H Medium

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli* and a variety of other bacteria.

H Top Agar

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g
Agar	7.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes that contain H agar.

Use: For the cultivation of *Escherichia coli* and a variety of other bacteria.

Halophile Medium

Composition per liter:

NaCl.....	30.0g
Agar	15.0g
Peptone	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Vibrio alginolyticus*, *Vibrio parahaemolyticus*, and *Vibrio proteolyticus*.

Halophilic Agar (HA)

Composition per liter:

NaCl.....	250.0g
MgSO ₄ ·7H ₂ O	25.0g
Agar	20.0g
Casamino acids	10.0g
Yeast extract.....	10.0g
Proteose peptone.....	5.0g
Trisodium citrate.....	3.0g
KCl.....	2.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Combine the ingredients with distilled water and heat to boiling to dissolve completely. Autoclave at 121°C for 15 min.

Use: For the isolation and cultivation of halophilic microorganisms from foods, such as *Pseudomonas* species and *Flavobacterium* species from fish and salted foods.

Halophilic Broth (HB)

Composition per liter:

NaCl.....	250.0g
MgSO ₄ ·7H ₂ O	25.0g
Casamino acids	10.0g
Yeast extract.....	10.0g
Proteose peptone.....	5.0g
Trisodium citrate.....	3.0g
KCl.....	2.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of halophilic microorganisms from foods, such as *Pseudomonas* species and *Flavobacterium* species from fish and salted foods.

Halophilic Nutrient Agar (LMG Medium 220)

Composition per liter:

NaCl.....	60.0g
Agar	15.0g

Casein peptone, tryptic digest.....	10.0g
Yeast extract.....	5.0g
Glucose	5.0g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of halophilic bacteria.

Halophilic Synthetic Medium

Composition per liter:

Glucose	0.1g
KNO ₃	0.05g
FePO ₄	0.01g
Artificial seawater.....	100.0mL

Artificial Seawater:

Composition per 100.0mL:

NaCl.....	2.4g
MgCl ₂ ·6H ₂ O	1.1g
Na ₂ SO ₄	0.4g
CaCl ₂ ·6H ₂ O	0.2g
KCl.....	0.07g
NaHCO ₃	0.02g
KBr	0.01g
SrCl ₂ ·6H ₂ O	4.0mg
H ₃ BO ₃	3.0mg
Na ₂ SiO ₃ ·9H ₂ O	0.5mg
NaF	0.3mg

Preparation of Artificial Seawater: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of halophilic bacteria.

Ham's F-10 Medium (BAM M58)

Composition per liter:

NaCl.....	7.4g
NaHCO ₃	1.2g
Glucose	1.1g
NaH ₂ PO ₄ ·H ₂ O.....	0.29g
KCl.....	0.28g
L-Arginine·HCl.....	0.21g
L-Glutamine	0.15g
MgSO ₄ ·7H ₂ O	0.15g
Sodium pyruvate.....	0.11g

KH ₂ PO ₄	0.08g
CaCl ₂ ·2H ₂ O.....	0.04g
L-Cystine·2HCl.....	0.04g
L-Histidine·HCl·H ₂ O.....	0.02g
L-Lysine·HCl.....	0.02g
L-Asparagine·H ₂ O.....	0.01g
L-Aspartic Acid.....	0.01g
L-Glutamic acid.....	0.01g
L-Leucine.....	0.01g
L-Proline.....	0.01g
L-Serine.....	0.01g
L-Alanine.....	8.9mg
Glycine.....	7.5mg
D-Phenylalanine.....	5.0mg
L-Methionine.....	4.5mg
Hypoxanthine.....	4.1mg
L-Threonine.....	3.6mg
L-Valine.....	3.5mg
L-Isoleucine.....	2.6mg
L-Tyrosine.....	1.8mg
Vitamin B ₁₂	1.4mg
Folic acid.....	1.3mg
Phenol Red.....	1.2mg
Thiamine·HCl.....	1.0mg
FeSO ₄ ·7H ₂ O.....	0.8mg
Choline chloride.....	0.7mg
D-Calcium pantothenate.....	0.7mg
Thymidine.....	0.7mg
Niacinamide.....	0.6mg
L-Tryptophan.....	0.6mg
Inositol.....	0.5mg
Riboflavin.....	0.4mg
Lipoic acid.....	0.2mg
Pyridoxine·HCl.....	0.2mg
ZnSO ₄ ·7H ₂ O.....	0.03mg
Biotin.....	0.02mg
CuSO ₄ ·5H ₂ O.....	3.0μg

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: For the growth of Y-1 cell cultures used in the mouse adrenal assay for heat-labile toxin of enterotoxigenic *Escherichia coli* and *Vibrio* species.

Heart Infusion Agar (HIA) (BAM M159)

Composition per liter:

Agar.....	15.0g
Beef heart, infusion from 500.0g.....	10.0g
Tryptose.....	10.0g
NaCl.....	5.0g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of a wide variety of fastidious microorganisms. For the cultivation and maintenance of *Bacillus cereus*, *Staphylococcus aureus*, and *Vibrio vulnificus* and *Vibrio cholerae*. It can also be used as a base for the preparation of blood agar in determining hemolytic reactions. When using for blood agar, reduce volume to 950.0mL to allow for addition of 50.0mL of defibrinated horse blood. Blood is added aseptically after autoclaving.

Heart Infusion Agar (HIA) (BAM M159)

Composition per liter:

Proteose Peptone.....	15.0g
Agar.....	12.0g
Yeast Extract.....	5.0g
NaCl.....	5.0g
Liver Digest.....	2.5g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of a wide variety of fastidious microorganisms. For the cultivation and maintenance of *Bacillus cereus*, *Staphylococcus aureus*, and *Vibrio vulnificus* and *Vibrio cholerae*. It can also be used as a base for the preparation of blood agar in determining hemolytic reactions. When using for blood agar, reduce volume to 950.0mL to allow for addition of 50.0mL of defibrinated horse blood. Blood is added aseptically after autoclaving.

Heart Infusion Agar (HIA) (BAM M60)

Composition per liter:

Agar.....	15.0g
Tryptose.....	10.0g

NaCl.....	5.0g
Beef heart, infusion from 500.0g.....	1.0L
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of a wide variety of fastidious microorganisms. For the cultivation and maintenance of *Bacillus cereus*, *Staphylococcus aureus*, and *Vibrio vulnificus* and *Vibrio cholerae*. It can also be used as a base for the preparation of blood agar in determining hemolytic reactions. When using for blood agar, reduce volume to 950.0mL to allow for addition of 50.0mL of defibrinated horse blood. Blood is added aseptically after autoclaving.

Heart Infusion Agar with NaCl (HIA with NaCl) (BAM M60)

Composition per liter:

NaCl.....	20.0g
Agar	15.0g
Tryptose	10.0g
NaCl.....	20.0g
Beef heart, infusion from 500.0g.....	1.0L
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of halophilic *Vibrio* spp.

Heart Infusion Broth (HI) (BAM M60)

Composition per liter:

Agar	15.0g
Tryptose	10.0g
NaCl.....	5.0g
Beef heart, infusion from 500.0g.....	1.0L
pH 7.4 ± 0.2 at 25°C	

Source: This medium without added NaCl is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of a wide variety of fastidious microorganisms. For the cultivation and maintenance of *Bacillus cereus*, *Staphylococcus aureus*, and *Vibrio vulnificus* and *Vibrio cholerae*. It can also be used as a base for the preparation of blood agar in determining hemolytic reactions. When using for blood broth, reduce volume to 950.0mL to allow for addition of 50.0mL of defibrinated horse blood. Blood is added aseptically after autoclaving.

Heart Infusion Broth with NaCl (HI with NaCl) (BAM M60)

Composition per liter:

NaCl.....	20.0g
Tryptose	10.0g
NaCl.....	20.0g
Beef heart, infusion from 500.0g.....	1.0L
pH 7.4 ± 0.2 at 25°C	

Source: This medium without added NaCl is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of halophilic *Vibrio* spp.

Hektoen Enteric Agar (BAM M61)

Composition per liter:

Agar	14.0g
Lactose	12.0g
Peptic digest of animal tissue	12.0g
Sucrose.....	12.0g
Bile salts.....	9.0g
NaCl.....	5.0g
Na ₂ S ₂ O ₃	5.0g
Yeast extract.....	3.0g
Salicin	2.0g
Ferric ammonium citrate.....	1.5g
Acid Fuchsin.....	0.1g
Bromthymol Blue	0.064g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until components are dissolved. Do not autoclave. Pour into sterile Petri dishes. Allow agar to solidify with the Petri dish covers partially off. Do not store more than 1 day.

Use: For the isolation and cultivation of Gram-negative enteric microorganisms based on lactose or sucrose fermentation and H₂S production. For the isolation and differentiation of *Salmonella* and *Shigella*. Bacteria that ferment lactose or sucrose appear as yellow to orange colonies. Bacteria that produce H₂S appear as colonies with black centers.

Hemorrhagic Coli Agar (HC Agar) (BAM M62)

Composition per liter:

Sorbitol.....	20.0g
Pancreatic digest of casein.....	20.0g
Agar	15.0g
NaCl.....	5.0g
Bile salts No. 3.....	1.12g
Bromcresol Purple	0.015g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation and cultivation of enterohemorrhagic *Escherichia coli* from food. For hemorrhagic colitis *E. coli* strains.

Hemorrhagic Coli Agar with MUG (HC Agar with MUG) (BAM M62)

Composition per liter:

Sorbitol.....	20.0g
Pancreatic digest of casein.....	20.0g
Agar	15.0g
NaCl.....	5.0g
Bile salts No. 3.....	1.12g

Bromcresol Purple	0.015g
MUG reagent	0.1g
pH 7.2 ± 0.2 at 25°C	

Source: MUG reagent is available from Hach Company, Loveland Colorado.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation and cultivation of enterohemorrhagic *Escherichia coli* from food. For hemorrhagic colitis *E. coli* strains.

HHD Medium

Composition per liter:

Agar	20.0g
Pancreatic digest of casein.....	10.0g
Casamino acids	3.0g
Fructose.....	2.5g
KH ₂ PO ₄	2.5g
Papaic digest of soybean meal.....	1.5g
Tween 80.....	1.0g
Yeast extract.....	1.0g
Bromcresol Green solution	20.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Green Solution:

Composition per 30.0mL:

Bromcresol Green.....	0.1g
NaOH (0.01N solution).....	30.0mL

Preparation of Bromcresol Green Solution:

Add Bromcresol Green to 30.0mL of NaOH solution. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Salmonella* species from foods.

HiCrome™ Aureus Agar Base (Aureus Agar Base)

**(HiCrome *Staphylococcus aureus* Agar)
(*Staphylococcus aureus* Agar, HiCrome)**

Composition per liter:

Agar	20.0g
Casein enzymic hydrolysate	12.0g
Sodium pyruvate.....	10.0g

Beef extract.....	6.0g
LiCl.....	5.0g
Yeast extract.....	5.0g
Pancreatic digest of of gelatin.....	3.0g
Chromogenic mixture.....	2.1g
Egg tellurite emulsion.....	50.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Caution: Lithium chloride is harmful. Avoid bodily contact and inhalation of vapors. On contact with skin, wash with plenty of water immediately.

Egg Yolk Tellurite Emulsion:

Composition per 100.0mL:

Sterile saline.....	64.0mL
Egg Yolk.....	30.0mL
Sterile potassium tellurite solution, 3.5%.....	6.0mL

Preparation of Medium: Add components, except egg yolk tellurite emulsion, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 50.0mL sterile egg yolk tellurite emulsion. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and enumeration of coagulase positive *Staphylococcus aureus*. Coagulase positive *S. aureus* gives brown black colonies whereas *S. epidermidis* gives yellow, slightly brownish, colonies.

HiCrome Coliform Agar (Coliform Agar, HiCrome)

Composition per liter:

Agar.....	12.0g
Sodium chloride.....	5.0g
Peptone, special.....	3.0g
K ₂ HPO ₄	3.0g
KH ₂ PO ₄	1.7g
Sodium pyruvate.....	1.0g
Tryptophan.....	1.0g
Chromogenic mixture.....	0.2g
Sodium lauryl sulfate.....	0.1g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the simultaneous detection of *Escherichia coli* and total coliforms in water and food samples.

Sodium lauryl sulfate inhibits gram-positive organisms. The chromogenic mixture contains two chromogenic substrates as Salmon-GAL and X-glucuronide. The enzyme β-D-galactosidase produced by coliforms cleaves Salmon-GAL, resulting in the salmon to red coloration of coliform colonies. The enzyme β-D-glucuronidase produced by *E. coli* cleaves X-glucuronide. *Escherichia coli* forms dark blue to violet colored colonies due to cleavage of both Salmon-GAL and X-glucuronide. The addition of tryptophan improves the indole reaction, thereby increasing detection reliability in combination with the two chromogens.

HiCrome Coliform Agar with Novobiocin (Coliform Agar with Novobiocin, HiCrome)

Composition per liter:

Agar.....	12.0g
Sodium chloride.....	5.0g
Peptone, special.....	3.0g
K ₂ HPO ₄	3.0g
KH ₂ PO ₄	1.7g
Sodium pyruvate.....	1.0g
Tryptophan.....	1.0g
Chromogenic mixture.....	0.2g
Sodium lauryl sulfate.....	0.1g
Novobiocin.....	5.0mg

pH 6.8 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the simultaneous detection of *Escherichia coli* and total coliforms in water and food samples when high numbers of Gram-positive bacteria may be present. Novobiocin and sodium lauryl sulfate inhibit Gram-positive organisms. The chromogenic mixture contains two chromogenic substrates as Salmon-GAL and X-glucuronide. The enzyme β-D-galactosidase produced by coliforms cleaves Salmon-GAL, resulting in the salmon to red coloration of coliform colonies. The enzyme β-D-glucuronidase produced by *E. coli* cleaves X-glucuronide. *Escherichia coli* forms dark blue to violet colored colonies due to cleavage of both Salmon-GAL and X-glucuronide. The addition of tryptophan improves the indole reaction, thereby increasing detection reliability in combination with the two chromogens.

HiCrome *E. coli* Agar A (*E. coli* Agar A, HiCrome)

Composition per liter:

Casein enzymic hydrolysate	14.0g
Agar	12.0g
Peptone, special	5.0g
NaCl.....	2.4g
Bile salts mixture	1.5g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g
X-Glucuronide	0.075g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Pour into sterile Petri dishes.

Use: For the detection and enumeration of *Escherichia coli* in foods without further confirmation on membrane filter or by indole reagent. The chromogenic agent X-glucuronide used in this medium helps to detect glucuronidase activity. *E. coli* cells absorb X-glucuronide and the intracellular glucuronidase splits the bond between the chromophore and the glucuronide. The released chromophore gives coloration to the colonies. Bile salts mixture inhibits gram-positive organisms.

HiCrome *E. coli* Agar B (*E. coli* Agar B, HiCrome)

Composition per liter:

Casein enzymic hydrolysate	20.0g
Agar	15.0g
Bile salts mixture	1.5g
X-Glucuronide	0.075g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the detection and enumeration of *Escherichia coli* in foods without further confirmation on membrane filter or by indole reagent. The chromogenic agent X-glucuronide used in this medium helps to detect glucuronidase activity. *E. coli* cells absorb X-glucuronide and the intracellular glucuronidase splits the bond between the chromophore

and the glucuronide. The released chromophore gives coloration to the colonies. Bile salts mixture inhibits gram-positive organisms.

HiCrome ECC Agar (ECC Agar, HiCrome)

Composition per liter:

Chromogenic mixture	20.3g
Agar	15.0g
Peptone, special	5.0g
NaCl.....	5.0g
Na ₂ HPO ₄	3.5g
Yeast extract.....	3.0g
Lactose	2.5g
KH ₂ PO ₄	1.5g
Neutral Red.....	0.03g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: A differential medium for presumptive identification of *E. coli* and other coliforms in food samples. The chromogenic mixture contains two chromogens as X-glucuronide and Salmon-GAL. X-glucuronide is cleaved by the enzyme β-glucuronidase produced by *E. coli*. Salmon-GAL is cleaved by the enzyme galactosidase produced by the majority of coliforms, including *E. coli*.

HiCrome ECC Selective Agar (ECC Selective Agar, HiCrome)

Composition per liter:

Agar	10.0g
Peptone, special	6.0g
Casein enzymic hydrolysate	3.3g
NaCl.....	2.0g
Sodium pyruvate.....	1.0g
Tryptophan.....	1.0g
Sorbitol	1.0g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g
Tergitol 7®.....	0.15g
Chromogenic mixture	0.43g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly and heat with frequent agitation until components are completely dissolved (approximately 35 min). Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Medium may show haziness.

Use: For detection of *Escherichia coli* and coliforms in water and food samples. Tergitol inhibits Gram-positive as well as some Gram-negative bacteria other than coliforms. The chromogenic mixture contains two chromogenic substrates as Salmon-GAL and X-glucuronide. The enzyme β -D-galactosidase produced by coliforms cleaves Salmon-GAL, resulting in the salmon to red coloration of coliform colonies. The enzyme β -D-glucuronidase produced by *E. coli* cleaves X-glucuronide. *E. coli* forms dark blue to violet colored colonies due to cleavage of both Salmon-GAL and X-glucuronide. The addition of tryptophan improves the indole reaction.

**HiCrome ECC Selective Agar
(ECC Selective Agar, HiCrome)**

Composition per liter:

Agar	10.0g
Peptone, special	6.0g
Casein enzymic hydrolysate	3.3g
NaCl	2.0g
Sodium pyruvate	1.0g
Tryptophan	1.0g
Sorbitol	1.0g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g
Tergitol 7	0.15g
Chromogenic mixture	0.43g
Cefulodin	5.0mg

pH 6.8 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until components are completely dissolved (approximately 35 min). Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Medium may show haziness.

Use: For detection of *Escherichia coli* and coliforms in water and food samples using pour plate or streak plate methods. Tergitol inhibits Gram-positive as well as some Gram-negative bacteria other than coliforms. The cefsulodin inhibits *Pseudomonas* and *Aeromonas* species. The chromogenic mixture contains two chromogenic substrates as Salmon-GAL and X-glucuronide. The enzyme β -D-galactosidase produced by coliforms cleaves Salmon-GAL, result-

ing in the salmon to red coloration of coliform colonies. The enzyme β -D-glucuronidase produced by *E. coli* cleaves X-glucuronide. *E. coli* forms dark blue to violet colored colonies due to cleavage of both Salmon-GAL and X-glucuronide. The addition of tryptophan improves the indole reaction.

**HiCrome ECC Selective Agar
(ECC Selective Agar, HiCrome)**

Composition per liter:

Agar	10.0g
Peptone, special	6.0g
Casein enzymic hydrolysate	3.3g
NaCl	2.0g
Sodium pyruvate	1.0g
Tryptophan	1.0g
Sorbitol	1.0g
Na ₂ HPO ₄	1.0g
NaH ₂ PO ₄	0.6g
Tergitol 7	0.15g
Chromogenic mixture	0.43g
Cefulodin	10.0mg

pH 6.8 ± 0.2 at 25°C

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly and heat with frequent agitation until components are completely dissolved (approximately 35 min). Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes. Medium may show haziness.

Use: For detection of *Escherichia coli* and coliforms in water and food samples using the membrane filter technique. Tergitol inhibits Gram-positive as well as some Gram-negative bacteria other than coliforms. The cefsulodin inhibits *Pseudomonas* and *Aeromonas* species. The chromogenic mixture contains two chromogenic substrates as Salmon-GAL and X-glucuronide. The enzyme β -D-galactosidase produced by coliforms cleaves Salmon-GAL, resulting in the salmon to red coloration of coliform colonies. The enzyme β -D-glucuronidase produced by *E. coli* cleaves X-glucuronide. *E. coli* forms dark blue to violet colored colonies due to cleavage of both Salmon-GAL and X-glucuronide. The addition of tryptophan improves the indole reaction.

HiCrome Enterococci Broth (Enterococci HiCrome Broth)

Composition per liter:

Peptone, special	10.0g
NaCl	5.0g
Polysorbate 80	2.0g
NaH ₂ PO ₄	1.25g
NaN ₃	0.3g
Chromogenic mixture	0.04g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Caution: Sodium azide is toxic. It also has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the rapid and easy identification and differentiation of enterococci. It contains a chromogenic substrate which aids in the detection of enterococci, especially in water samples.

HiCrome *Listeria* Agar Base, Modified (*Listeria* HiCrome Agar Base, Modified)

Composition per liter:

Peptone, special	23.0g
Agar	13.0g
Rhamnose	10.0g
Chromogenic mixture	5.13g
LiCl	5.0g
Meat extract	5.0g
NaCl	5.0g
Yeast extract	1.0g
Phenol Red	0.12g
pH 7.3 ± 0.2 at 25°C	

Caution: Lithium chloride is harmful. Avoid bodily contact and inhalation of vapors. On contact with skin, wash with plenty of water immediately.

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: A selective and differential agar medium recommended for rapid and direct identification of *Listeria* species, specifically *Listeria monocytogenes*.

HiCrome MacConkey-Sorbitol Agar MacConkey-Sorbitol Agar, HiCrome

Composition per liter:

Casein enzymic hydrolysate	17.0g
Agar	13.5g
Sorbitol	10.0g
NaCl	5.0g
Proteose peptone	3.0g
Bile salts mixture	1.5g
5-Bromo-4-chloro-3-indolyl-β-D-glucuronide sodium salt	0.1g
Neutral Red	0.03g
Crystal Violet	0.001g
pH 7.1 ± 0.3 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Mix to completely dissolve components. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes.

Use: For the direct isolation and differentiation of *E. coli* O157:H7-strains from foodstuffs. Recommended for selective isolation of *Escherichia coli* O157:H7 from food and animal feeding stuffs. The medium contains sorbitol instead of lactose. Enteropathogenic strains of *Escherichia coli* O157:H7 ferment lactose but do not ferment sorbitol and hence produce colorless colonies. Sorbitol fermenting strains of *Escherichia coli* produce pink-red colonies. The red color is due to production of acid from sorbitol, absorption of neutral red, and a subsequent color change of the dye when pH of the medium falls below 6.8. The chromogenic indicator is added to detect the presence of an enzyme β-D-glucuronidase. Strains of *Escherichia coli* possessing β-D-glucuronidase appear as blue colored colonies on the medium. Enteropathogenic strains of *Escherichia coli* O157 do not possess β-D-glucuronidase activity and thus produce colorless colonies. *Escherichia coli* fermenting sorbitol and possessing β-D-glucuronidase activity produce purple colored colonies. Most of the gram-positive organisms are inhibited by crystal violet and bile salts.

HiCrome MacConkey-Sorbitol Agar with Tellurite-Cefixime Supplement (MacConkey-Sorbitol Agar with Tellurite-Cefixime Supplement)

Composition per 1004mL:

Casein enzymic hydrolysate	17.0g
Agar	13.5g

Sorbitol.....	10.0g
NaCl.....	5.0g
Proteose peptone.....	3.0g
Bile salts mixture.....	1.5g
5-Bromo-4-chloro-3-indolyl- β -D-glucuronide sodium salt.....	0.1g
Neutral Red.....	0.03g
Crystal Violet.....	0.001g
Tellurite-cefixime supplement.....	4.0mL
pH 7.1 ± 0.3 at 25°C	

Tellurite-Cefixime Supplement:**Composition per 4.0mL:**

K ₂ TeO ₃	5.0mg
Cefiximine.....	0.1mg

Preparation of Tellurite-Cefixime Supplement: Add components to 4.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Source: This medium is available from Fluka, Sigma-Aldrich. Tellurite-cefixime supplement is available from Oxoid.

Preparation of Medium: Add components, except tellurite-cefixime supplement, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Mix to completely dissolve components. Do not autoclave. Cool to 50°C. Add 4.0mL sterile tellurite-cefixime supplement. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the direct isolation and differentiation of *E. coli* O157:H7-strains from foodstuffs. Recommended for selective isolation of *Escherichia coli* O157:H7 from food and animal feeding stuffs. The medium contains sorbitol instead of lactose. Enteropathogenic strains of *Escherichia coli* O157:H7 ferment lactose but do not ferment sorbitol and hence produce colorless colonies. Sorbitol fermenting strains of *Escherichia coli* produce pink-red colonies. The red color is due to production of acid from sorbitol, absorption of neutral red, and a subsequent color change of the dye when pH of the medium falls below 6.8. The chromogenic indicator is added to detect the presence of an enzyme β -D-glucuronidase. Strains of *Escherichia coli* possessing β -D-glucuronidase appear as blue colored colonies on the medium. Enteropathogenic strains of *Escherichia coli* O157 do not possess β -D-glucuronidase activity and thus produce colorless colonies. *Escherichia coli* fermenting sorbitol and possessing β -D-glucuronidase activity produce purple colored colonies. Most of the gram-positive organisms are inhibited by crystal violet and bile salts. Addition of Tellurite-Cefixime Supplement makes the medium selective. Potassium Tel-

lurite selects the serogroups O157 from other *E. coli* serogroups and inhibits *Aeromonas* species and *Providencia* species. Cefixime inhibits *Proteus* species.

**HiCrome M-CP Agar Base
(M-CP HiCrome Agar Base)
(Membrane *Clostridium perfringens*
HiCrome Agar Base)**

Composition per liter:

Tryptose.....	30.0g
Yeast extract.....	20.0g
Agar.....	15.0g
Sucrose.....	5.0g
L-Cysteine-HCl-H ₂ O.....	1.0g
MgSO ₄ ·7H ₂ O.....	0.1g
FeCl ₃ ·6H ₂ O.....	0.09g
Indoxyl- β -D-glucoside.....	0.06g
Bromocresol Purple.....	0.04g
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the detection of *Clostridium perfringens*. Recommended by the Directive of the Council of the European Union 98/83/EC for isolation and enumeration of *C. perfringens* from water samples using membrane filtration technique.

**HiCrome MS.O157 Agar
(MS.O157 Agar HiCrome)**

Composition per liter:

Agar.....	12.0g
Peptone, special.....	10.0g
Sorbitol.....	4.0g
Bile salt mixture.....	1.0g
Chromogenic mixture.....	0.731g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes.

Use: For the simultaneous detection of *Escherichia coli*, *Escherichia coli* O157:H7, and coliforms in water and food samples. *Escherichia coli* O157 gives

colorless colonies because of non-fermentation of sorbitol and absence of β -glucuronidase activity, whereas other strains of *Escherichia coli* having β -glucuronidase activity and fermenting sorbitol appear as steel blue colored colonies. Some non-*Escherichia coli* O157 may have some colony color.

**HiCrome MS.O157 Agar
with Tellurite
(MS.O157 Agar
with Tellurite, HiCrome)**

Composition 1000.25mL:

Agar	12.0g
Peptone, special	10.0g
Sorbitol.....	4.0g
Bile salt mixture.....	1.0g
Chromogenic mixture	0.731g
Tellurite solution	0.25mL
pH 6.8 ± 0.2 at 25°C	

Tellurite Solution:

Composition per 10.0mL:

K_2TeO_3	0.1g

Preparation of Tellurite Solution: Add components to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components, except tellurite solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 45°C. Aseptically add 0.25mL sterile tellurite solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the simultaneous detection of *Escherichia coli*, *Escherichia coli* O157:H7, and coliforms in water and food samples. *Escherichia coli* O157:H7 gives colorless colonies because of non-fermentation of sorbitol and absence of β -glucuronidase activity, whereas other strains of *Escherichia coli* having β -glucuronidase activity and fermenting sorbitol appear as steel blue colored colonies. Addition of tellurite makes the medium much more specific and selective.

**HiCrome OGYE Agar Base
(OGYE Agar Base HiCrome)
(Oxytetracyclin Glucose
Yeast Extract Agar HiCrome)**

Composition per liter:

Glucose	20.0g
Agar	12.0g

Yeast extract	4.0g
Chromogenic mixture	1.1g
Oxytetracycline selective supplement	10.0mL
pH 7.0 ± 0.2 at 25°C	

Oxytetracycline Selective Supplement:

Composition per 10.0mL:

Oxytetracycline.....	0.1g

Preparation of Oxytetracycline Selective Supplement: Add 0.1g oxytetracycline to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components, except oxytetracycline selective supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL sterile oxytetracycline selective supplement. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the rapid isolation of yeasts and molds from milk and milk products. Oxytetracycline makes the medium more selective by inhibiting the growth of lactobacilli. *Aspergillus niger* appears as light blue colored colonies with black spores due to presence of chromogenic mixture, *Candida albicans* shows green colored colonies and *Saccharomyces cerevisiae* gives colorless colonies.

**HiCrome Rapid Coliform Broth
(Coliform Rapid HiCrome Broth)
(Rapid Coliform HiCrome Broth)**

Composition per liter:

Peptone, special	5.0g
NaCl.....	5.0g
Na_2HPO_4	2.7g
KH_2PO_4	2.0g
Sorbitol	1.0g
Sodium lauryl sulfate.....	0.1g
IPTG.....	0.1g
Chromogenic substrate	0.08g
Fluorogenic substrate.....	0.05g
pH 6.8 ± 0.3 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the detection and conformation of *Escherichia coli* and coliforms on the basis of enzyme substrate reaction from water samples, using a combination of chromogenic and fluorogenic substrate.

HiCrome Rapid Enterococci Agar
(Enterococci Rapid HiCrome Agar)
(Rapid Enterococci HiCrome Agar)

Composition per liter:

Agar	15.0g
Peptone special	10.0g
NaCl	5.0g
Polysorbate 80	2.0g
Na ₂ HPO ₄	1.25g
NaN ₃	0.3g
Chromogenic mixture	0.06g
pH 7.5 ± 0.2 at 25°C	

Caution: Sodium azide is toxic. It also has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the rapid and easy identification and differentiation of enterococci. It contains a chromogenic substrate, which aids in the detection of enterococci, especially from water samples.

HiCrome Salmonella Agar
(Salmonella Agar, HiCrome)

Composition per liter:

Agar	13.0g
Peptic digest of animal tissue	6.0g
Chromogenic mixture	5.4g
Yeast extract	2.5g
Bile salt mixture	1.0g
pH 7.7 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Mix to completely dissolve components. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes.

Use: A selective chromogenic medium used for the isolation and differentiation of *Salmonella* species from coliforms in food and water. *E. coli* and *Salmonella* are easily distinguishable due to the colony characteristics. *Salmonella* give light purple colonies with a halo. *E. coli* has a characteristic blue color. Other organisms give colorless colonies. The characteristic light purple and blue color is due to the chromogenic mixture.

HL Agar

Composition per plate:

Columbia agar base	10.0mL
Columbia blood top agar	5.0mL
pH 7.3 ± 0.2 at 25°C	

Columbia Agar Base:

Composition per liter:

Agar	13.5g
Pancreatic digest of casein	12.0g
NaCl	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	3.0g
Yeast extract	3.0g
Cornstarch	1.0g

Preparation of Columbia Agar Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Columbia Blood Top Agar:

Composition per liter:

Agar	13.5g
Pancreatic digest of casein	12.0g
NaCl	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	3.0g
Yeast extract	3.0g
Cornstarch	1.0g
Horse blood, defibrinated	50.0mL

Preparation of Columbia Blood Top Agar:

Add components, except horse blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile horse blood. Mix thoroughly.

Preparation of Medium: Pour cooled, sterile Columbia agar base into sterile Petri dishes in 10.0mL volumes. Allow agar to solidify. Pour 5.0mL of cooled, sterile Columbia blood top agar over Columbia agar base that has solidified but is still warm.

Use: For the cultivation of *Listeria monocytogenes*.

Horie Arabinose Ethyl Violet Broth (HAEB)

Composition per liter:

NaCl.....	30.0g
Peptone.....	5.0g
Beef extract.....	3.0g
Bromthymol Blue	0.03g
Ethyl Violet.....	1.0mg
Arabinose solution	100.0mL
pH 9.0 ± 0.2 at 25°C	

Arabinose Solution:

Composition per 100.0mL:

Arabinose.....	5.0g

Preparation of Arabinose Solution: Add arabinose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except arabinose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 9.0. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile arabinose solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Preparation of Medium: Dissolve all ingredients, except arabinose, in 900.0mL of distilled water. Adjust the pH to 9.0 ± 0.2 and sterilize at 121°C for 15 min. Cool and aseptically add 100.0mL of a filter-sterilized 5% arabinose solution in distilled water. After mixing, aseptically dispense the medium into sterile tubes or bottles.

Use: For the cultivation of *Vibrio* species from foods.

HP 101 Halophile Medium

Composition per liter:

NaCl.....	100.0g
Agar	20.0g
Peptone.....	10.0g
MgSO ₄ ·7H ₂ O	4.3g
NaNO ₃	2.0g
Yeast extract.....	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Pseudomonas* species.

HP Medium

Composition per liter:

Pancreatic digest of soybean meal.....	20.0g
Beef extract.....	10.0g
Yeast extract.....	6.0g
Ammonium citrate	5.0g
Tween 80.....	0.5g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
FeSO ₄ ·7H ₂ O.....	0.04g
Glucose solution	10.0mL
Tetracycline solution.....	10.0mL

Glucose Solution:

Composition per 100.0mL:

Glucose	10.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Tetracycline Solution:

Composition per 100.0mL:

Tetracycline.....	10.0g

Preparation of Tetracycline Solution: Add tetracycline to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution and tetracycline solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile glucose solution and tetracycline solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and enumeration of *Leuconostoc* species.

HPC Agar (Heterotrophic Plate Count Agar) (m-HPC Agar)

Composition per liter:

Gelatin.....	25.0g
Pancreatic digest of gelatin.....	20.0g
Agar	15.0g
Glycerol	10.0mL
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available from BD Diagnostic Systems.

Preparation of Medium: Add components, except glycerol, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Add glycerol. Mix thoroughly.

Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the the cultivation and enumeration of microorganisms by the membrane filter method and heterotrophic plate count technique.

Hugh-Leifson's Glucose Broth (HLGB) (BAM M63)

Composition per liter:

NaCl	30.0g
Glucose	10.0g
Agar	3.0g
Peptone	2.0g
Yeast extract	0.5g
Bromeresol Purple	0.015g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Adjust pH to 7.4. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to ferment glucose. Bacteria that ferment glucose turn the medium yellow.

HY Agar for *Flavobacterium*

Composition per liter:

Agar	8.0g
Glutamic acid	5.0g
K ₂ HPO ₄	0.1g
MgSO ₄ ·7H ₂ O	0.1g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Glutamic acid may be replaced by 1.0g of folic acid if desired. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Flavobacterium* species.

HY Medium for *Flavobacterium*

Composition per liter:

Glutamic acid	5.0g
K ₂ HPO ₄	0.1g
MgSO ₄ ·7H ₂ O	0.1g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Glutamic acid may be replaced by 1.0g of folic acid

if desired. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Flavobacterium* species.

HYA Agar

Composition per liter:

Agar	15.0g
Proteose peptone No. 3	10.0g
Beef extract	1.0g
Lactose solution	10.0mL
Galactose solution	10.0mL
Glucose solution	10.0mL
pH 6.8 ± 0.2 at 25°C	

Lactose Solution:

Composition per 10.0mL:

Lactose	5.0g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Galactose Solution:

Composition per 10.0mL:

Galactose	2.5g

Preparation of Galactose Solution: Add galactose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Glucose Solution:

Composition per 10.0mL:

Glucose	2.5g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except lactose solution, galactose solution, and glucose solution—to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.8. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile lactose solution, galactose solution, and glucose solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of acidogenic microorganisms, especially *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, from foods.

Hydroxybenzoate Medium

Composition per 1002.0mL:

Solution A	920.0mL
Solution B	50.0mL
Solution C	1.0mL
Solution D	1.0mL

Solution E	10.0mL
Solution F	10.0mL
Solution G	10.0mL

pH 7.2–7.5 at 25°C

Solution A:

Composition per 920.0mL:

NaCl	1.0g
KCl	0.5g
MgCl ₂ ·6H ₂ O	0.4g
NH ₄ Cl	0.25g
KH ₂ PO ₄	0.2g
CaCl ₂ ·2H ₂ O	0.15g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Solution B:

Composition per 50.0mL:

NaHCO ₃	2.5g

Preparation of Solution B: Add NaHCO₃ to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Solution C (Trace Elements Solution SL-10):

Composition per liter:

FeCl ₂ ·4H ₂ O	1.5g
CoCl ₂ ·6H ₂ O	190.0mg
MnCl ₂ ·4H ₂ O	100.0mg
ZnCl ₂	70.0mg
Na ₂ MoO ₄ ·2H ₂ O	36.0mg
NiCl ₂ ·6H ₂ O	24.0mg
H ₃ BO ₃	6.0mg
CuCl ₂ ·2H ₂ O	2.0mg
HCl (25% solution)	10.0mL

Preparation of Solution C (Trace Elements Solution SL-10): Add FeCl₂·4H₂O to 10.0mL of HCl solution. Mix thoroughly. Add distilled/deionized water and bring volume to 1.0L. Add remaining components. Mix thoroughly. Gas under 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Solution D:

Composition per 10.0mL:

NaOH	5.0mg
Na ₂ WO ₄ ·2H ₂ O	40.0μg
Na ₂ SeO ₃ ·5H ₂ O	30.0μg

Preparation of Solution D: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Solution E (Vitamin Solution):

Composition per liter:

Pyridoxine·HCl	10.0mg
Calcium DL-pantothenate	5.0mg
Lipoic acid	5.0mg
Nicotinic acid	5.0mg
p-Aminobenzoic acid	5.0mg
Riboflavin	5.0mg
Thiamine·HCl	5.0mg
Biotin	2.0mg
Folic acid	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Solution E (Vitamin Solution):

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Solution F:

Composition per 10.0mL:

Sodium dihydroxybenzoate	0.4g

Preparation of Solution F: Add sodium dihydroxybenzoate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Sparge with 80% N₂ + 20% CO₂.

Solution G:

Composition per 10.0mL:

Na ₂ S·9H ₂ O	0.36g

Preparation of Solution G: Add Na₂S·9H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Sparge with 80% N₂ + 20% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Prepare and dispense medium under 80% N₂ + 20% CO₂. Aseptically and anaerobically combine 920.0mL of sterile solution A, 50.0mL of sterile solution B, 1.0mL of sterile solution C, 1.0mL of sterile solution D, 10.0mL of sterile solution E, 10.0mL of sterile solution F, and 10.0mL of sterile solution G. Mix thoroughly. Aseptically and anaerobically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Clostridium* species.

Hydroxybenzoic Acid Medium

Composition per liter:

Agar	15.0g
K ₂ HPO ₄ ·3H ₂ O	4.25g
NH ₄ Cl	2.0g
4-Hydroxybenzoic acid	1.0g
NaH ₂ PO ₄ ·H ₂ O	1.0g
MgSO ₄ ·7H ₂ O	0.2g
Nitrilotriacetic acid	0.1g
FeSO ₄ ·7H ₂ O	0.012g

MnSO ₄ ·H ₂ O	3.0mg
ZnSO ₄ ·7H ₂ O	3.0mg
CoSO ₄	1.0mg
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add 4-hydroxybenzoic acid and nitrilotriacetic acid to approximately 600.0mL of distilled/deionized water. Adjust pH to 8.0 with concentrated NaOH. Add remaining components. Mix thoroughly. Readjust pH to 7.2. Bring volume to 1.0L with distilled/deionized water. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Bacillus* species.

IE Medium

Composition per 1011.0mL:

Agar	15.0g
Peptone.....	5.0g
Yeast extract.....	1.0g
Basal salts solution.....	1.0L
Lactose solution	10.0g
Trace elements solution	1.0mL

Basal Salts Solution:

Composition per liter:

MgSO ₄	0.5g
Phosphate solution	20.0mL
(NH ₄)SO ₄ (36% solution)	5.0mL

Phosphate Solution:

Composition per liter:

K ₂ HPO ₄	95.0g
NaH ₂ PO ₄ ·2H ₂ O.....	78.0g

Preparation of Phosphate Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Basal Salts Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.8–7.0.

Lactose Solution:

Composition per 10.0mL:

Lactose	2.5g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Warm to 50°–55°C.

Trace Elements Solution:

Composition per liter:

Disodium EDTA	0.5g
FeSO ₄ ·7H ₂ O.....	0.2g
H ₃ BO ₃	0.03g
CoCl ₂ ·6H ₂ O	0.02g

ZnSO ₄ ·7H ₂ O	0.01g
MnCl ₂ ·4H ₂ O	3.0mg
Na ₂ MoO ₄ ·2H ₂ O	3.0mg
NiCl ₂ ·6H ₂ O.....	2.0mg
CaCl ₂ ·2H ₂ O	1.0mg

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Combine components, except lactose solution and trace elements solution. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 10.0mL of sterile lactose solution and 1.0mL of sterile trace elements solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Bacillus* species.

Imidazole Utilization Medium

Composition per liter:

Imidazole	5.0g
KH ₂ PO ₄	0.5g
MgSO ₄ ·7H ₂ O	0.5g
CaCl ₂	3.0mg
FeSO ₄ ·7H ₂ O.....	3.0mg
Molybdenum solution.....	1.0mL
Trace element solution.....	1.0mL

pH 6.0 ± 0.2 at 25°C

Molybdenum Solution:

Composition per 18.0mL:

Na ₂ MoO ₄ ·2H ₂ O	0.5mg

Preparation of Molybdenum Solution: Add components to distilled/deionized water and bring volume to 18.0mL. Mix thoroughly. Filter sterilize.

Trace Element Solution:

Composition per 18.0mL:

H ₃ BO ₃	11.0mg
MnCl ₂ ·4H ₂ O	7.0mg
Al ₂ (SO ₄) ₃ ·18 H ₂ O	1.94mg
Co(NO ₃) ₂ ·6H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O.....	1.0mg
NiSO ₄ ·6H ₂ O.....	1.0mg
ZnSO ₄ ·H ₂ O	0.62mg
KBr	0.5mg
KI	0.5mg
LiCl	0.5mg
SnCl ₂ ·2H ₂ O.....	0.5mg

Preparation of Trace Element Solution: Add components to distilled/deionized water and bring volume to 18.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except molybdenum solution and trace element solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 1.0mL of molybdenum solution and 1.0mL of trace element solution. Mix thoroughly. Adjust pH to 6.0 with phosphoric acid. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Pseudomonas* species.

Indole Medium (BAM M64)

Composition per 200.0mL:

K ₂ HPO ₄	3.13g
L-Tryptophan.....	1.0g
NaCl.....	1.0g
KH ₂ PO ₄	0.27g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of microorganisms by means of indole production from the tryptophan test.

Indole Medium, CDC (BAM M65)

Composition per liter:

Pancreatic digest of casein.....	20.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add pancreatic digest of casein to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of microorganisms by means of the indole test.

Indole Nitrite Medium (Trypticase™ Nitrate Broth) (Tryptic Nitrite Medium) (BAM M66)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Na ₂ HPO ₄	2.0g
Agar.....	1.0g

Glucose.....	1.0g
KNO ₃	1.0g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling with frequent agitation. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the identification of microorganisms by means of the nitrate reduction and indole tests.

Infection Medium (IM)

Composition per 100.0mL:

Pancreatic digest of gelatin.....	0.05g
Bile salts No. 3.....	0.05g
Brain heart, solids from infusion.....	0.02g
Peptic digest of animal tissue.....	0.02g
NaCl.....	0.017g
Glucose.....	0.01g
Na ₂ HPO ₄	8.0mg
Earle's balanced salts solution.....	80.0mL
Fetal bovine serum, heat inactivated (2 hr at 55°C).....	20.0mL
pH 7.4 ± 0.2 at 25°C	

Earle's Balanced Salts Solution:

Composition per liter:

NaCl.....	6.8g
NaHCO ₃	2.2g
Glucose.....	1.0g
KCl.....	0.4g
CaCl ₂ ·2H ₂ O.....	0.265g
MgSO ₄ ·7H ₂ O.....	0.2g
NaH ₂ PO ₄ ·H ₂ O.....	0.14g

Preparation of Earle's Balanced Salts Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Combine components. Mix thoroughly. Filter sterilize. Store at 4°–10°C.

Use: For the screening of *Escherichia coli* for pathogenicity using the HeLa cell test for invasiveness.

Infusion Broth

Composition per liter:

Pancreatic digest of casein.....	13.0g
NaCl.....	5.0g

Yeast extract.....	5.0g
Heart muscle, solids from infusion	2.0g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C .

Use: For the cultivation of a wide variety of microorganisms.

Inositol Brilliant Green Bile Salts Agar (IBB Agar) (*Plesiomonas* Differential Agar)

Composition per liter:

Agar	15.0g
meso-Inositol.....	10.0g
Proteose peptone.....	10.0g
Bile salts No. 3.....	8.5g
Meat extract	5.0g
NaCL.....	5.0g
Neutral Red (2% solution)	1.25mL
Brilliant Green (0.1% solution)	0.33mL
pH 7.2 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation of *Aeromonas* and *Plesiomonas* species.

Inositol Gelatin Deep

Composition per liter:

Gelatin.....	120.0g
Inositol	10.0g
Na_2HPO_4	5.0g
Yeast extract.....	5.0g
Phenol Red.....	0.05g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.4. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 10 psi pressure– 115°C .

Use: For the cultivation of *Plesiomonas shigelloides* from foods.

Irgasan Ticarcillin Chlorate Broth (ITC Broth) (BAM M67)

Composition per liter:

$\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$	60.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
KClO_4	1.0g
Yeast extract.....	1.0g
Malachite Green (0.2% solution).....	5.0mL
Irgasan solution.....	1.0mL
Ticarcillin solution	1.0mL
pH 7.6 ± 0.2 at 25°C	

Irgasan Solution:

Composition per 10.0mL:

Irgasan (triclosan)	1.0mg

Preparation of Irgasan Solution: Add Irgasan to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Ticarcillin Solution:

Composition per 10.0mL:

Ticarcillin.....	1.0mg

Preparation of Ticarcillin Solution: Add ticarcillin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except Irgasan solution and ticarcillin solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 45° – 50°C . Adjust to pH 7.6. Aseptically add 1.0mL of Irgasan solution and 1.0mL of ticarcillin solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the selective isolation and cultivation of *Yersinia* species.

Iron Agar, Lyngby (Lyngby Iron Agar)

Composition per liter:

Peptone	20.0g
Agar	12.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Yeast extract.....	3.0g
L-Cysteine	0.6g
Ferric citrate.....	0.3g
$\text{Na}_2\text{S}_2\text{O}_3$	0.3g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of H₂S-producing bacteria and total counts of heterotrophic bacteria from fish and fish products.

Iron Milk Medium

Composition per liter:

Iron filings.....	1.0g
Whole milk	1.0L
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add iron filings, which may be small balls of steel wool, to whole milk and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of lactic acid bacteria. For the cultivation and differentiation of *Clostridium* species. The medium turns black if H₂S is produced. The medium turns red if acid is produced from milk carbohydrate fermentation. Acid and gas production are characteristic of *Clostridium perfringens* and *Clostridium butyricum*.

Iron Milk Medium, Modified (BAM M68)

Composition per 1050.0mL:

Whole milk, fresh	1.0L
FeSO ₄ ·7H ₂ O	1.0g

Preparation of Medium: Add FeSO₄·7H₂O to distilled/deionized water and bring volume to 50.0mL. Add slowly to 1.0L of whole milk. Mix thoroughly. Distribute into tubes in 11.0mL volumes. Autoclave for 12 min at 13 psi pressure–118°C.

Use: For the cultivation and enumeration of *Clostridium perfringens* in foods.

IsoVitaleX Enrichment

Composition per liter:

Glucose	100.0g
L-Cysteine·HCl.....	25.9g
L-Glutamine.....	10.0g
Adenine	1.0g
Thiamine pyrophosphate.....	0.1g
Vitamin B ₁₂	0.1g
Guanine·HCl	0.03g
Fe(NO ₃) ₃ ·9H ₂ O	0.02g

p-Aminobenzoic acid.....	0.013g
Thiamine·HCl	0.003g

Preparation of IsoVitaleX Enrichment: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: As a nutrient supplement for the isolation and cultivation of fastidious microorganisms.

J Agar

Composition per liter:

Agar	20.0g
Yeast extract.....	15.0g
Pancreatic digest of casein.....	5.0g
K ₂ HPO ₄	3.0g
Glucose solution	10.0mL
pH 7.3–7.5 at 25°C	

Glucose Solution:

Composition per 10.0mL:

Glucose	2.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of *Bacillus* species and *Sporolactobacillus* species.

J Broth

Composition per liter:

Yeast extract.....	15.0g
Pancreatic digest of casein.....	5.0g
pH 7.3–7.5 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.3–7.5. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus* species and *Sporolactobacillus* species for performing the Voges-Proskauer test.

Jordan's Tartrate Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	10.0g
Sodium potassium tartrate	10.0g

NaCl.....	5.0g
Phenol Red.....	0.024g
pH 7.7 ± 0.3 at 25°C	

Source: This medium is available as a prepared medium in tubes from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.7. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation and identification of members of the Enterobacteriaceae, especially *Salmonella* species, based upon the ability to utilize tartrate. Utilization of tartrate turns the medium yellow. *Salmonella enteritidis* utilizes tartrate. *Salmonella paratyphi* A does not utilize tartrate.

Kado's Agar

Composition per liter:

Agar	15.0g
Sucrose.....	10.0g
Pancreatic digest of casein.....	8.0g
Yeast extract.....	4.0g
K ₂ HPO ₄	2.0g
MgSO ₄ ·7H ₂ O	30.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Lactococcus lactis* subspecies *hordniae*.

Kanamycin Esculin Azide Agar

Composition per liter:

Pancreatic digest of casein.....	20.0g
Agar	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Esculin	1.0g
Sodium citrate	1.0g
Ferric ammonium citrate.....	0.5g
NaN ₃	0.15g
Kanamycin sulfate solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Kanamycin Sulfate Solution:

Composition per 10.0mL:

Kanamycin sulfate	20.0mg

Preparation of Kanamycin Sulfate Solution:

Add kanamycin sulfate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation of enterococci from foods.

Kanamycin Esculin Azide Broth

Composition per liter:

Pancreatic digest of casein.....	20.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Esculin	1.0g
Sodium citrate	1.0g
Ferric ammonium citrate.....	0.5g
NaN ₃	0.15g
Kanamycin sulfate solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Kanamycin Sulfate Solution:

Composition per 10.0mL:

Kanamycin sulfate	0.02g

Preparation of Kanamycin Sulfate Solution:

Add kanamycin sulfate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation of enterococci from foods.

KC Bottom Agar

Composition per liter:

Agar	10.0g
Pancreatic digest of casein.....	10.0g
KCl.....	2.5g
NaCl.....	2.5g
CaCl ₂ solution.....	1.0mL

CaCl₂ Solution:

Composition per 10.0mL:

CaCl ₂ ·2H ₂ O	1.47g

Preparation of CaCl₂ Solution: Add 1.47g of CaCl₂·2H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except CaCl₂ solution, to distilled/deionized water and bring volume to 999.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 1.0mL of CaCl₂ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

KC Broth

Composition per liter:

Pancreatic digest of casein	10.0g
KCl	5.0g
CaCl ₂ solution	0.5mL

CaCl₂ Solution:

Composition per 10.0mL:

CaCl ₂ ·2H ₂ O	1.47g

Preparation of CaCl₂ Solution: Add 1.47g of CaCl₂·2H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except CaCl₂ solution, to distilled/deionized water and bring volume to 999.5mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 0.5mL of CaCl₂ solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the cultivation of *Escherichia coli*.

KC Top Agar

Composition per liter:

Pancreatic digest of casein	10.0g
Agar	8.0g
NaCl	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

KCN Broth

Composition per liter:

Na ₂ HPO ₄	5.64g
NaCl	5.0g

Peptone	3.0g
KH ₂ PO ₄	0.225g
KCN (0.5% solution)	15.0mL
pH 7.6 ± 0.2 at 25°C	

Caution: Cyanide is toxic.

Preparation of Medium: Add components, except KCN solution, to distilled/deionized water and bring volume to 985.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add KCN solution. Mix thoroughly. Aseptically distribute into sterile tubes. Stopper immediately.

Use: For the differentiation of Enterobacteriaceae based upon growth in the presence of potassium cyanide.

KF *Streptococcus* Agar

Composition per liter:

Agar	20.0g
Maltose	20.0g
Proteose peptone	10.0g
Sodium glycerophosphate	10.0g
Yeast extract	10.0g
NaCl	5.5g
Lactose	1.0g
NaN ₃	0.4g
Bromcresol Purple	0.015g
2,3,5-Triphenyltetrazolium chloride solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

2,3,5-Triphenyltetrazolium Chloride Solution:

Composition per 10.0mL:

2,3,5-Triphenyltetrazolium chloride	0.1g

Preparation of 2,3,5-Triphenyltetrazolium Chloride Solution: Add 2,3,5-triphenyltetrazolium chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except 2,3,5-triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2,3,5-triphenyltetrazolium chloride solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and enumeration of enterococci.

KF Streptococcus Agar**Composition per liter:**

Agar	20.0g
Maltose.....	20.0g
Sodium glycerophosphate.....	10.0g
Yeast extract.....	10.0g
NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
Lactose	1.0g
NaN ₃	0.4g
2,3,5-Triphenyltetrazolium chloride solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

2,3,5-Triphenyltetrazolium Chloride Solution:**Composition per 10.0mL:**

2,3,5-Triphenyltetrazolium chloride	0.1g

Preparation of 2,3,5-Triphenyltetrazolium

Chloride Solution: Add 2,3,5-triphenyltetrazolium chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except 2,3,5-triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2,3,5-triphenyltetrazolium chloride solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective cultivation and enumeration of fecal streptococci.

KF Streptococcus Broth**Composition per liter:**

Maltose.....	20.0g
Sodium glycerophosphate.....	10.0g
Yeast extract.....	10.0g
NaCl.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
Lactose	1.0g
Na ₂ CO ₃	0.636g
NaN ₃	0.4g

Phenol Red.....	0.018g
2,3,5-Triphenyltetrazolium chloride solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except 2,3,5-triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2,3,5-triphenyltetrazolium chloride solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the selective cultivation of fecal streptococci.

**Kim-Goepfert Agar
(KG Agar)****Composition per 1001.0mL:**

Solution A	900.0mL
Egg yolk emulsion, 50%.....	100.0mL
Polymyxin B solution	1.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Solution A:**Composition per 900.0mL:**

Agar	18.0g
Peptone	1.0g
Yeast extract.....	0.5g
Phenol Red.....	0.025g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 6.8. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Polymyxin B Solution:**Composition per 5.0mL:**

Polymyxin B sulfate..... 500,000U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 900.0mL of cooled, sterile solution A, aseptically add 100.0mL of sterile egg yolk emulsion, 50%, and 1.0mL of sterile polymyxin B solution. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry in the dark at 30°C for 24 hr before using.

Use: For the cultivation and differentiation of *Bacillus cereus*.

King's B Medium (BAM M69)

Composition per liter:

Proteose peptone No. 3 20.0g
Agar 150.0g
K₂HPO₄, anhydrous 1.5g
MgSO₄·7H₂O 1.5g
Glycerol 10.0mL

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the nonselective isolation, cultivation, and pigment production of *Pseudomonas* species.

King's Medium A**Composition per liter:**

Proteose peptone 20.0g
Agar 15.0g
Glycerol 10.0g
K₂SO₄ 10.0g
MgCl₂·6H₂O 3.5g

pH 7.2–7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the nonselective isolation, cultivation, and pigment production of *Pseudomonas*.

King's Medium B**Composition per liter:**

Agar 20.0g
Proteose peptone No. 3 20.0g
K₂HPO₄, anhydrous 1.5g
MgSO₄·7H₂O 1.5g
Glycerol 15.0mL

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the nonselective isolation, cultivation, and pigment production of *Pseudomonas* species.

King's O/F Basal Medium (Oxidation-Fermentation Medium, King's) (King's OF Medium) (BAM M70)

Composition per liter:**Base:**

Agar 3.0g
Pancreatic digest of casein 2.0g
Carbohydrate solution 100.0mL
Phenol Red (1.5% solution) 2.0mL
pH to 7.3 ± 0.2

Carbohydrate Solution:**Composition per 100.0mL:**

Carbohydrate 10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For differentiating bacteria based upon determining the oxidative and fermentative metabolism of carbohydrates. Bacteria that ferment the carbohydrate turn the medium yellow.

Kleb Agar (m-Kleb Agar)

Composition per liter:

Agar 15.0g

Proteose peptone No. 3	10.0g
NaCl	5.0g
Adonitol	5.0g
Beef extract	1.0g
Aniline Blue	0.1g
Sodium lauryl sulfate	0.1g
Phenol Red	0.025g
Ethanol (95% solution)	20.0mL
Carbenicillin solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Carbenicillin Solution:**Composition per 10.0mL:**

Carbenicillin	0.05g

Preparation of Carbenicillin Solution: Add carbenicillin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except ethanol and carbenicillin solution, to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 20.0mL of ethanol and 10.0mL of carbenicillin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the enumeration of bacteria from waters.

Kligler Iron Agar**Composition per liter:**

Peptone	20.0g
Agar	12.0g
Lactose	10.0g
NaCl	5.0g
Beef extract	3.0g
Yeast extract	3.0g
Glucose	1.0g
Ferric citrate	0.3g
Na ₂ S ₂ O ₃	0.3g
Phenol Red	0.05g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation and identification of Enterobacteriaceae based upon sugar fermentation and hydrogen sulfide production. Sugar fermentation is

indicated by the medium turning yellow. H₂S production results in the medium turning black.

Kligler Iron Agar**Composition per liter:**

Agar	15.0g
Lactose	10.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g
Ferric ammonium citrate	0.5g
Na ₂ S ₂ O ₃	0.5g
Phenol Red	0.025g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation and identification of Enterobacteriaceae based upon sugar fermentation and hydrogen sulfide production. Sugar fermentation is indicated by the medium turning yellow. H₂S production results in the medium turning black.

**Kligler Iron Agar
(FDA M71)****Composition per liter:**

Agar	15.0g
Lactose	20.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g
Ferric ammonium citrate	0.5g
Na ₂ S ₂ O ₃	0.5g
Phenol Red	0.025g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation and identification of Enterobacteriaceae based upon sugar fermentation and hydrogen sulfide production. Sugar fermentation is

indicated by the medium turning yellow. H₂S production results in the medium turning black.

Kligler Iron Agar (BAM M71)

Composition per liter:

Lactose	20.0g
Polypeptone peptone	20.0g
Agar	15.0g
Peptic digest of animal tissue.....	10.0g
NaCl	5.0g
Glucose	1.0g
Ferric ammonium citrate.....	0.5g
Na ₂ S ₂ O ₃	0.5g
Phenol Red	0.025g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation and identification of Enterobacteriaceae based upon sugar fermentation and hydrogen sulfide production. Sugar fermentation is indicated by the medium turning yellow. H₂S production results in the medium turning black.

Kligler Iron Agar with NaCl (BAM M71)

Composition per liter:

NaCl	25.0g
Lactose	20.0g
Polypeptone peptone	20.0g
Agar	15.0g
Peptic digest of animal tissue.....	10.0g
NaCl	5.0g
Glucose	1.0g
Ferric ammonium citrate.....	0.5g
Na ₂ S ₂ O ₃	0.5g
Phenol Red	0.025g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation and identification of *Vibrio* spp. based upon sugar fermentation and hydrogen sulfide production. Sugar fermentation is indicated by the medium turning yellow. H₂S production results in the medium turning black.

KM Agar (Kempfer-McKay Agar)

Composition per liter:

Agar	15.0g
Milk, nonfat	10.0g
Glucose	5.0g
Milk protein hydrolysate.....	2.5g
Solution 1	10.0mL
Solution 2	10.0mL

pH 6.6 ± 0.2 at 25°C

Solution 1:

Composition per 100.0mL:

K ₃ Fe(CN) ₆	10.0g

Preparation of Solution 1: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Solution 2:

Composition per 40.0mL:

Ferric citrate	1.0g
Sodium citrate	1.0g

Preparation of Solution 2: Add components to distilled/deionized water and bring volume to 40.0mL. Mix thoroughly. Filter sterilize.

Caution: Cyanide is toxic.

Preparation of Medium: Add components, except solution 1 and solution 2, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.6. Autoclave for 12 min at 10 psi pressure–115°C. Cool to 45°–50°C. Aseptically add sterile solution 1 and solution 2. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry in the dark at 30°C for 24 hr before using.

Use: For the isolation and cultivation of acidogenic microorganisms from foods. For the differentiation of citrate-fermenting lactic bacteria, such as *Lactobacillus lactis* subspecies *diacetylactis*, from the noncitrate-fermenting *Lactobacillus lactis* subspecies *cremoris*.

Korthof Medium

Composition per 1088.0mL:

NaCl	1.4g
Na ₂ HPO ₄ ·2H ₂ O	0.88g
Peptone	0.8g
KH ₂ PO ₄	0.24g
CaCl ₂	0.04g
KCl	0.04g
NaHCO ₃	0.02g
Rabbit serum, inactivated	80.0mL
Rabbit hemoglobin solution	8.0mL

pH 7.2 ± 0.2 at 25°C

Rabbit Hemoglobin Solution:**Composition** per 20.0mL:

Rabbit blood clot..... 10.0mL

Preparation of Rabbit Hemoglobin Solution:

Add rabbit blood clot to 10.0mL of distilled/deionized water. Lyse the clot by freezing and thawing.

Preparation of Medium: Add components, except rabbit serum and rabbit hemoglobin solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 25°C. Filter through Whatman #1 filter paper. Distribute into flasks in 100.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 8.0mL of rabbit serum and 0.8mL of rabbit hemoglobin solution to each flask. Mix thoroughly.

Use: For the cultivation of *Leptospira* species.

Korthof Medium, Modified**Composition** per liter:

NaCl.....1.4g
 Na₂HPO₄·2H₂O.....0.88g
 Peptone.....0.8g
 KH₂PO₄.....0.24g
 CaCl₂.....0.04g
 KCl.....0.04g
 NaHCO₃.....0.02g
 Rabbit serum, heat inactivated at 56°C..... 100.0mL
 pH 7.2–7.6 at 25°C

Preparation of Medium: Add components, except rabbit serum, to distilled/deionized water and bring volume to 900.0L. Mix thoroughly. Gently heat and bring to boiling. Boil for 20 min. Cool overnight at 4°C. Filter through Whatman #2 filter paper. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–56°C. Aseptically add 100.0mL of rabbit serum. Mix thoroughly.

Use: For the cultivation of *Leptospira* species.

**Koser Citrate Broth
(BAM M72)****Composition** per liter:

Sodium citrate.....3.0g
 NaNH₄HPO₄·4H₂O.....1.5g
 KH₂PO₄.....1.0g
 MgSO₄·7H₂O.....0.2g
 pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute

into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of *Escherichia coli* and *Enterobacter aerogenes* based on citrate utilization.

Kranep Agar Base**Composition** per liter:

KSCN.....25.5g
 Agar.....18.3g
 Sodium pyruvate.....8.2g
 Pancreatic digest of gelatin.....6.1g
 LiCl.....5.1g
 Mannitol.....5.1g
 Beef extract.....3.7g
 NaN₃.....0.05g
 Cycloheximide.....0.041g
 Egg yolk emulsion..... 100.0mL
 pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Egg Yolk Emulsion:**Composition:**

Chicken egg yolks..... 11
 Whole chicken egg..... 1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and enumeration of staphylococci from foods.

Kreb's Yeast Lactate Medium**Composition** per liter:

Yeast extract.....10.0g
 Na₂HPO₄·2H₂O.....3.0g
 KH₂PO₄.....1.0g
 Sodium lactate solution..... 40.0mL
 pH 7.0 ± 0.2 at 25°C

Sodium Lactate Solution:**Composition** per 100.0mL:

Sodium lactate..... 70.0g

Preparation of Sodium Lactate Solution: Add sodium lactate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sodium lactate solution, to distilled/deionized water and bring volume to 960.0mL. Mix thoroughly. Bring pH to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 40.0mL of sterile sodium lactate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Propionibacterium acidipropionici*, *Propionibacterium freudenreichii*, *Propionibacterium intermedium*, *Propionibacterium jensenii*, *Propionibacterium* species, and *Propionibacterium thoenii*.

Kunkee Medium**Composition** per 1000.88mL:

Pancreatic digest of casein..... 20.0g
Glucose 5.0g
Peptone..... 5.0g
Yeast extract..... 5.0g
Filtered tomato juice 250.0mL
Ethanol (96% solution) 176.0mL
Tween 80..... 0.5mL

pH 5.5 ± 0.2 at 25°C

Preparation of Medium: Add components, except ethanol, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature. Aseptically add 0.88mL of ethanol solution to each tube containing 5.0mL of medium.

Use: For the cultivation of *Lactobacillus fructivorans*, which has been isolated from spoiled sake.

KYE Agar**Composition** per liter:

Agar 15.0g
NaNO₃..... 2.5g
KH₂PO₄..... 1.0g
Yeast extract..... 1.0g
MgSO₄·7H₂O 0.3g
CaCl₂·6H₂O..... 0.15g
NaCl..... 0.1g
FeCl₃ 10.0mg

pH 10.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix

thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of alkaliphilic bacteria.

**L Agar
(Luria Agar)****Composition** per liter:

Agar 15.0g
Pancreatic digest of casein..... 10.0g
Yeast extract..... 5.0g
NaCl..... 0.5g
Glucose 20.0mL

pH 7.0 ± 0.2 at 25°C

Glucose Solution:**Composition** per 100.0mL:

Glucose 10.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Bring pH to 7.0. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 20.0mL of sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Escherichia coli*.

**L Medium
(ATCC Medium 1154)****Composition** per liter:

Pancreatic digest of casein..... 10.0g
NaCl..... 5.0g
Yeast extract..... 5.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli*.

L Medium for Salmonella**Composition** per liter:

Pancreatic digest of casein..... 10.0g
NaCl..... 5.0g

Yeast extract.....	5.0g
Glucose	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C .

Use: For the cultivation and maintenance of *Salmonella*.

L15 Medium, Modified Leibovitz (BAM M73)

Composition per liter:

NaCl.....	8.0g
DL-Threonine.....	0.6g
Sodium pyruvate.....	0.6g
DL-Alanine.....	0.5g
L-Arginine, free base.....	0.5g
KCl.....	0.4g
L-Asparagine·H ₂ O.....	0.3g
L-Histidine, free base.....	0.3g
L-Glutamine.....	0.3g
L-Isoleucine.....	0.3g
L-Phenylalanine.....	0.3g
L-Tyrosine.....	0.3g
DL-Methionine.....	0.2g
DL-Valine.....	0.2g
Glycine.....	0.2g
L-Serine.....	0.2g
Na ₂ HPO ₄ , anhydrous.....	0.2g
CaCl ₂ , anhydrous.....	0.1g
L-Cysteine, free base.....	0.1g
L-Leucine·HCl.....	0.1g
MgCl ₂ , anhydrous.....	0.094g
D-Galactose.....	0.09g
KH ₂ PO ₄	0.06g
L-Tryptophan.....	0.02g
Phenol Red.....	0.01g
<i>i</i> -Inositol.....	2.0mg
Choline chloride.....	1.0mg
D-Calcium pantothenate.....	1.0mg
Folic acid.....	1.0mg
Nicotinamide.....	1.0mg
Pyridoxine·HCl.....	1.0mg
Thiamine monophosphate·2H ₂ O.....	1.0mg
Riboflavin-5-phosphate.....	0.1mg
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Store at 5°C .

Use: For the cultivation of oysters used for the growth of enteroviruses.

Lab-Lemco Agar

Composition per liter:

Agar.....	15.0g
Peptone.....	5.0g
Lab-lemco meat extract.....	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of heterotrophic microorganisms.

Lab-Lemco Broth

Composition per liter:

Peptone.....	5.0g
Lab-lemco meat extract.....	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C .

Use: For the cultivation of a variety of heterotrophic microorganisms.

Lactic Acid Bacteria Broth

Composition per liter:

Sodium acetate.....	12.0g
Pancreatic digest of casein.....	10.0g
Yeast autolysate.....	5.0g
Glucose.....	10.0g
Solution A.....	5.0mL
Solution B.....	5.0mL
pH 5.1 ± 0.2 at 25°C	

Source: Yeast autolysate is available from Oxoid.

Solution A:

Composition per 100.0mL:

K ₂ HPO ₄	10.0g
KH ₂ PO ₄	10.0g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Solution B:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O.....	4.0g
FeSO ₄ ·7H ₂ O.....	0.2g
MnSO ₄ ·H ₂ O.....	0.2g
NaCl.....	0.2g

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus malefermentans*.

Lactic Acid Bacteria Medium

Composition per liter:

Agar	15.0g
Glucose	10.0g
KH ₂ PO ₄	5.0g
Peptone	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
Diammonium hydrogen citrate	2.0g
Tween 80	1.0g
Sorbic acid	0.5g
MgSO ₄ ·7H ₂ O	0.5g
MnSO ₄ ·4H ₂ O	0.2g
FeSO ₄ ·7H ₂ O	0.05g

pH 5.3–5.4 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.3–5.4. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of lactic acid bacteria from wine.

Lactic Agar for Yogurt Bacteria, Modified

Composition per liter:

Elliker agar	1.0L
Milk solution	70.0mL

pH 6.8 ± 0.1 at 25°C

Elliker Agar:

Composition per liter:

Pancreatic digest of casein	20.0g
Agar	15.0g
Yeast extract	10.0g
Gelatin	4.0g
Glucose	3.0g
Ascorbic acid	2.5g
Lactose	2.5g
NaCl	2.5g
Sodium acetate	2.5g
Sucrose	2.5g

Preparation of Elliker Agar: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Milk Solution:

Composition per 100.0mL:

Nonfat dry milk solids	11.0g

Preparation of Milk Solution: Add nonfat dry milk solids to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 12 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add 70.0mL of sterile milk solution to 1.0L of cooled, sterile Elliker agar. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry at 28°–30°C for 18–24 hr.

Use: For the cultivation of acidogenic microorganisms, especially *Lactobacillus* species and lactic streptococci, from foods.

Lactic Bacteria Broth

Composition per liter:

Sodium acetate	12.0g
Glucose	10.0g
Pancreatic digest of casein	10.0g
Yeast autolysate	5.0g
Solution A	5.0mL
Solution B	5.0mL

pH 5.1–5.3 at 25°C

Solution A:

Composition per 100.0mL:

K ₂ HPO ₄	10.0g
KH ₂ PO ₄	10.0g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Solution B:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O	4.0g
FeSO ₄	0.2g
MnSO ₄ ·5H ₂ O	0.2g
NaCl	0.2g

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus buchneri*, *Lactobacillus delbrueckii*, and *Pediococcus damnosus*.

Lactic Streak Agar

Composition per liter:

Agar	15.0g
Sodium carboxymethylcellulose	10.0g
Calcium citrate	10.0g
Beef extract	5.0g
Papaic digest of soybean meal	5.0g
Polypeptone	5.0g
Yeast extract	5.0g
Lactose	1.5g
L-Arginine-HCl	1.5g
Bromocresol Purple (0.1% solution)	20.0mL
pH 6.0 ± 0.1 at 25°C	

Preparation of Medium: Add components—except Bromocresol Purple solution, calcium citrate, and sodium carboxymethylcellulose—to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. In a blender, add the calcium citrate and sodium carboxymethylcellulose to 200.0mL of distilled/deionized water. Blend until mixed. Add the 200.0mL of citrate/carboxymethylcellulose solution to the hot agar solution. Mix thoroughly. Adjust pH to 6.0. Distribute into flasks in 100.0mL volumes. Autoclave for 10 min at 10 psi pressure–115°C. Cool to 45°–50°C. Aseptically add 2.0mL of sterile Bromocresol Purple to each flask. Mix thoroughly. Pour into sterile Petri dishes. Allow plates to dry at 37°C for 1 hr before use.

Use: For the differentiation of lactic streptococci. Bacteria that produce acid from lactose appear as yellow colonies.

Lactobacilli Agar, AOAC (Lactobacilli Agar, Association of Official Analytical Chemists)

Composition per liter:

Milk, peptonized	15.0g
Agar	10.0g
Glucose	10.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	1.0g
Tomato juice	100.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the cultivation and maintenance of stock cultures of *Lactobacillus casei* ATCC 7469, *Lactobacillus fermentum* ATCC 9338, *Lactobacillus leichmannii* ATCC 4797, and *Lactobacillus viridescens* ATCC 12706 used in the microbiological assay of vitamins.

Lactobacilli Broth, AOAC (Lactobacilli Broth, Association of Official Analytical Chemists)

Composition per liter:

Milk, peptonized	15.0g
Glucose	10.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	1.0g
Tomato juice	100.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and preparation of inocula of stock cultures of *Lactobacillus casei* ATCC 7469, *Lactobacillus fermentum* ATCC 9338, *Lactobacillus leichmannii* ATCC 4797, and *Lactobacillus viridescens* ATCC 12706 used in the microbiological assay of vitamins.

Lactobacilli MRS Broth (Lactobacilli deMan- Rogosa-Sharpe Broth)

Composition per liter:

Glucose	20.0g
Beef extract	10.0g
Peptone	10.0g
Sodium acetate	5.0g
Yeast extract	5.0g
Ammonium citrate	2.0g
Na ₂ HPO ₄	2.0g
Tween 80	1.0g

MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·5H ₂ O	0.05g
pH 6.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus* species. Also used for the cultivation and maintenance of *Aerococcus viridans*, *Bifidobacterium coryneforme*, *Lactococcus plantarum*, *Leuconostoc* species, *Pectinatus cerevisiiphilus*, *Pediococcus* species, and *Sporolactobacillus inulinus*.

Lactobacilli MRS Broth with Cysteine

Composition per liter:

Glucose	20.0g
Beef extract	10.0g
Peptone	10.0g
Sodium acetate	5.0g
Yeast extract	5.0g
L-Cysteine	2.0g
Ammonium citrate	2.0g
Na ₂ HPO ₄	2.0g
Tween 80	1.0g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·5H ₂ O	0.05g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus* species, including *L. ruminis* and *L. vitulinus*.

Lactobacilli MRS Broth with Ethanol

Composition per liter:

Glucose	20.0g
Beef extract	10.0g
Peptone	10.0g
Sodium acetate	5.0g
Yeast extract	5.0g
L-Cysteine	2.0g
Ammonium citrate	2.0g
Na ₂ HPO ₄	2.0g
Tween 80	1.0g
MgSO ₄ ·7H ₂ O	0.1g

MnSO ₄ ·5H ₂ O	0.05g
Ethanol (95% solution)	100.0mL
pH 5.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except ethanol, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Filter sterilize ethanol. Aseptically add 100.0mL of sterile ethanol. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Lactobacillus* species.

Lactobacillus bulgaricus Agar (LB Agar)

Composition per 900.0mL:

Agar	20.0g
Glucose	20.0g
Beef extract	10.0g
Pancreatic digest of casein	10.0g
Yeast extract	5.0g
K ₂ HPO ₄	2.0g
Tween 80	1.0g
Acetate buffer	80.0mL
Tomato juice, filtered	40.0mL
pH 6.8 ± 0.2 at 25°C	

Acetate Buffer:

Composition per liter:

Sodium acetate	113.55g
Acetic acid	10.0mL

Preparation of Acetate Buffer: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add components, except acetate buffer, to distilled/deionized water and bring volume to 820.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.8. Add 80.0mL of acetate buffer. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and enumeration of *Lactobacillus bulgaricus* from foods.

Lactobacillus Chloramphenicol Agar 1

Composition per liter:

Milk, peptonized	15.0g
Agar	10.0g
Glucose	10.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	1.0g

Chloramphenicol.....	0.3g
Tomato juice	100.0mL
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the cultivation of *Lactobacillus casei*.

***Lactobacillus* Chloramphenicol Broth 1**

Composition per liter:

Milk, peptonized	15.0g
Glucose	10.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	1.0g
Chloramphenicol.....	0.3g
Tomato juice	100.0mL
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus casei*.

***Lactobacillus* Chloramphenicol Medium 2**

Composition per liter:

Glucose	18.5g
Agar	13.5g
Pancreatic digest of gelatin	10.0g
Beef extract.....	8.0g
Yeast extract.....	4.0g
Sodium acetate.....	3.0g
K ₂ HPO ₄	2.0g
Ammonium citrate.....	2.0g
Polysorbate 80.....	1.0g
MgSO ₄ ·7H ₂ O	0.2g
Chloramphenicol.....	0.1g
MnSO ₄ ·4H ₂ O	0.05g
pH 6.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Lactobacillus casei* and *Pediococcus acidilactici*.

***Lactobacillus* Heteroferm Screen Agar (MRS, Modified)**

Composition per liter:

Glucose	20.0g
Agar	15.0g
Proteose peptone No. 3	10.0g
Sodium acetate.....	5.0g
Yeast extract.....	5.0g
2-Phenylethyl alcohol.....	3.0g
Ammonium citrate.....	2.0g
K ₂ HPO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·H ₂ O	0.05g
Bromcresol Green.....	0.04g
Cycloheximide.....	4.0mg
Tween 80.....	1.0mL
pH 5.5 ± 0.01 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Adjust pH to 5.5 with glacial acetic acid. Pour into sterile Petri dishes.

Use: For the isolation and cultivation of *Lactobacillus* species from salad dressings.

***Lactobacillus* Heteroferm Screen Broth (MRS, Modified)**

Composition per liter:

Glucose	20.0g
Proteose peptone No. 3	10.0g
Sodium acetate.....	5.0g
Yeast extract.....	5.0g
2-Phenylethyl alcohol.....	3.0g
Ammonium citrate.....	2.0g
K ₂ HPO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·H ₂ O	0.05g
Bromcresol Green.....	0.04g
Cycloheximide.....	4.0mg
Tween 80.....	1.0mL
pH 4.3 ± 0.01 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 4.3 with concentrated HCl. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Lactobacillus* species from foods.

Lactobacillus Medium (ATCC Medium 169)

Composition per liter:

Glucose	10.0g
Proteose peptone	7.5g
Yeast extract	7.5g
KH ₂ PO ₄	2.0g
L-Cysteine·HCl·H ₂ O	1.0g
Tween 80	0.1g
Tomato juice, filtered	100.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus casei*.

Lactobacillus Medium (ATCC Medium 1006)

Composition per liter:

Agar	20.0g
Pancreatic digest of casein	10.0g
Glucose	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	3.0g
K ₂ HPO ₄	3.0g
Tryptose	3.0g
Sodium acetate	1.0g
L-Cysteine·HCl·H ₂ O	0.2g
Salt solution R	5.0mL
Tween 80	1.0mL
pH 6.3 ± 0.2 at 25°C	

Salt Solution R:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O	11.5g
MnSO ₄ ·2H ₂ O	2.4g
FeSO ₄ ·7H ₂ O	0.68g

Preparation of Salt Solution R: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Store at 4°C.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus delbrueckii* and *Lactobacillus jensenii*.

Lactobacillus Medium

Composition per liter:

Glucose	20.0g
Sodium acetate	20.0g
Tryptone	10.0g
Yeast extract	5.0g
Beef extract	2.0g
K ₂ HPO ₄	0.5g
KH ₂ PO ₄	0.5g
MgSO ₄ ·7H ₂ O	200.0mg
FeSO ₄ ·7H ₂ O	10.0mg
MnSO ₄ ·H ₂ O	7.5mg
Ethanol	40.0mL
Mevalonic acid solution	10.0mL
Tween 80	1.0mL
pH 5.2 ± 0.2 at 25°C	

Mevalonic Acid Solution:

Composition per 10.0mL:

DL-Mevalonic acid	30.0mg

Preparation of Mevalonic Acid Solution: Add DL-mevalonic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except mevalonic acid solution and ethanol, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 40.0mL of filter-sterilized ethanol and 10.0mL of sterile mevalonic acid solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Lactobacillus homohiochii*.

Lactobacillus Medium (Lactobacillus Sake Medium)

Composition per liter:

Agar	13.0g
Yeast extract	5.0g
Liver extract concentrate	0.2g
Sake	700.0mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus fructivorans* and *Lactobacillus homohiochii*.

***Lactobacillus* Medium II**
(DSMZ Medium 93)**Composition per liter:**

Glucose	20.0g
Na-acetate	20.0g
Agar	15.0g
Pancreatic digest of casein	10.0g
Yeast extract	5.0g
Meat extract	2.0g
KH ₂ PO ₄	0.5g
K ₂ HPO ₄	0.5g
MgSO ₄ ·7H ₂ O	0.2g
DL-mevalonic acid	30.0mg
FeSO ₄ ·7H ₂ O	10.0mg
MnSO ₄ ·2H ₂ O	7.5mg
Ethanol	40.0mL
Tween 80	1.0mL

pH 5.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus fructivorans* and *Lactobacillus homohiochii*.

Lactobacillus* Medium III*Composition per liter:**

Pancreatic digest of casein	10.0g
Fructose	7.0g
Glucose	7.0g
Maltose	7.0g
Meat extract	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
K ₂ HPO ₄ ·3H ₂ O	2.6g
Diammonium citrate	2.0g
Sodium gluconate	2.0g
L-Cysteine·HCl	0.5g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·4H ₂ O	0.05g
Tween 80	1.0mL

pH 6.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.3. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus* species.

***Lactobacillus* Medium IV**
(DSMZ Medium 859)**Composition per liter:**

Tryptone	10.0g
Yeast extract	5.0g
Na-acetate·3H ₂ O	5.0g
Glucose	4.0g
Maltose	4.0g
Meat extract	3.0g
K ₂ HPO ₄ ·3H ₂ O	2.6g
(NH ₄) ₂ citrate	2.0g
Cysteine·HCl·H ₂ O	0.5g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·4H ₂ O	0.05g
Tween 80	1.0mL

pH 6.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus panis*.

Lactobacillus* 8664 Medium*Composition per liter:**

Maltose	20.0g
Peptone	10.0g
Yeast extract	10.0g
Glucose	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus brevis*.

Lactobacillus* Orotic Acid Medium*Composition per liter:**

Milk, peptonized	15.0g
Glucose	10.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	1.0g
Orotic acid	25.0mg
D-Pantothenic acid	0.2mg
Tomato juice	100.0mL
L-Cysteine·HCl·H ₂ O solution	7.5mL

pH 6.8 ± 0.2 at 25°C

L-Cysteine·HCl·H₂O Solution:**Composition per 10.0mL:**

L-Cysteine·HCl·H ₂ O	0.15g

Preparation of L-Cysteine·HCl·H₂O Solution: Add L-cysteine·HCl·H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into bottles in 20.0mL volumes. Add 0.15mL of L-cysteine-HCl-H₂O solution to each bottle containing 20.0mL of medium. Autoclave for 15 min at 15 psi pressure–121°C. Screw caps tightly to maintain reduced conditions.

Use: For the cultivation of *Lactobacillus helveticus*.

***Lactobacillus* Selection Oxgall Agar (LBST[™] Oxgall Agar)**

Composition per liter:

Sodium acetate-3H ₂ O	25.0g
Glucose	20.0g
Agar	15.0g
Pancreatic digest of casein	10.0g
KH ₂ PO ₄	6.0g
Yeast extract	5.0g
Ammonium citrate	2.0g
Oxgall	1.5g
Polysorbate 80	1.0g
MgSO ₄	0.575g
FeSO ₄	0.034g
MnSO ₄	0.12g
Acetic acid, glacial	1.32mL
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components, except acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and enumeration of lactobacilli.

Lactose Broth (BAM M74)

Composition per liter:

Lactose	5.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Cool

broth quickly to 25°C. For testing water samples with 10.0mL volumes, prepare medium double strength.

Use: For the detection of lactose-fermenting, Gram-negative coliforms, as a preenrichment broth for *Salmonella* species, and in the study of lactose fermentation of bacteria in general.

Lactose Egg Yolk Milk Agar

Composition per 1206.0mL:

Lactose	12.0g
Agar	1.0g
Columbia blood agar base	800.0mL
Skim milk	150.0mL
Egg yolk emulsion, 50%	36.0mL
Inhibitor solution	20.0mL
Neutral Red (1% solution)	3.25mL
pH 7.0 ± 0.2 at 25°C	

Columbia Blood Agar Base:

Composition per 800.0mL:

Agar	15.0g
Pantone	10.0g
Bitone	10.0g
NaCl	5.0g
Tryptic digest of beef heart	3.0g
Cornstarch	1.0g

Preparation of Columbia Blood Agar Base:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling.

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Inhibitor Solution:

Composition per 20.0mL:

Neomycin sulfate	0.18g
NaN ₃	0.24g

Preparation of Inhibitor Solution: Add neomycin sulfate and NaN₃ to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Combine Columbia blood agar base, lactose, agar, and Neutral Red and bring volume to 1.0L. Adjust pH to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 55°C. Filter sterilize skim milk. To 1.0L of cooled, sterile agar mixture, aseptically add 150.0mL of sterile skim milk, 36.0mL of sterile egg yolk emulsion, 50%, and 20.0mL of sterile inhibitor solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Clostridium* species.

Lactose Gelatin Medium (BAM M75)

Composition per liter:

Gelatin.....	120.0g
Tryptose	15.0g
Lactose	10.0g
Yeast extract.....	10.0g
Phenol Red (1.0% in ethanol solution)	5.0mL
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add tryptose, yeast extract, and lactose to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly. Add gelatin to distilled/deionized water and bring volume to 590.0mL. Gently heat gelatin solution while stirring and bring to 50°–60°C. Add Phenol Red. Mix the two solutions together. Distribute into tubes in 10.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C. If medium is not used in 8 hr, deoxygenate by heating to 50°–70°C for 2–3 hr prior to inoculation.

Use: For the cultivation of *Clostridium perfringens*.

Lactose Ricinoleate Broth

Composition per liter:

Lactose	10.0g
Peptone.....	5.0g
Sodium ricinoleate	1.0g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective cultivation of members of the Enterobacteriaceae.

Lauryl Sulfate Broth (m-Lauryl Sulfate Broth)

Composition per liter:

Peptone.....	39.0g
Lactose	30.0g
Yeast extract.....	6.0g

Sodium lauryl sulfate	1.0g
Phenol Red.....	0.2g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into bottles or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and enumeration of coliform bacteria, especially *Escherichia coli*, in water by the membrane filter method.

Lauryl Sulfate Broth (Lauryl Tryptose Broth)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
Sodium lauryl sulfate	0.1g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Cool broth quickly to 25°C. For testing water samples with 10.0mL volumes, prepare medium double strength.

Use: For the detection of coliform bacteria, including for the enumeration of coliform bacteria by the multiple-tube fermentation technique.

Lauryl Sulfate Broth, Fluorocult (Fluorocult Lauryl Sulfate Broth)

Composition per liter:

Tryptose	20.0g
Lactose	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
L-tryptophan.....	1.g
Sodium lauryl sulfate	0.1g
4-Methylumbelliferyl-β-D-glucuronide.....	0.1g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix

thoroughly. Gently heat and bring to boiling. Cool. Distribute into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C. The prepared broth is clear and yellowish-brown.

Use: For the detection of *E. coli* in milk. The medium complies with the German-DIN-Norm 10183 for the examination of milk, with the regulations acc. to § 35 LMBG (01.00/54) for the examination of food, and according to ISO/DIS 11886-2.2 (1994) for milk and milk products. The lauryl sulfate largely inhibits the growth of undesirable microbial flora. The presence of *E. coli* is indicated by fluorescence under a long wavelength UV lamp. A positive indole reaction and gas formation due to fermentation of lactose confirm the results.

Lauryl Sulfate Broth with MUG

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
Sodium lauryl sulfate.....	0.1g
4-Methylumbelliferyl- β-D-glucuronide (MUG).....	0.05g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Cool broth quickly to 25°C. For testing water samples with 10.0mL volumes, prepare medium double strength.

Use: For the detection of *Escherichia coli* in food samples by a fluorogenic procedure.

Lauryl Tryptose Broth (Lauryl Sulfate Broth)

(LST Broth)
(BAM M76)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
Sodium lauryl sulfate.....	0.1g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Cool broth quickly to 25°C. For testing water samples with 10.0mL volumes, prepare medium double strength.

Use: For the detection of coliform bacteria in a variety of specimens. Also, for the enumeration of coliform bacteria by the multiple-tube fermentation technique.

Lauryl Tryptose Broth with MUG (Lauryl Sulfate Broth with MUG) (LST-MUG) (BAM M77)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
Sodium lauryl sulfate.....	0.1g
4-Methylumbelliferyl- β-D-glucuronide (MUG).....	0.05g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Cool broth quickly to 25°C. For testing water samples with 10.0mL volumes, prepare medium double strength.

Use: For the detection of *Escherichia coli* in water and food samples by a fluorogenic procedure.

Lauryl Tryptose Mannitol Broth with Tryptophan

Composition per liter:

Pancreatic digest of casein.....	20.0g
Lactose.....	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.75g
KH ₂ PO ₄	2.75g
Sodium lauryl sulfate.....	0.1g
L-Tryptophan.....	0.2g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes containing an inverted Durham tube in 10.0mL volumes. Autoclave for 10 min at 10 psi pressure–115°C. Cool broth quickly to 25°C.

Use: For the detection of *Escherichia coli*.

LB Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
NaCl	5.0g
Yeast extract	5.0g
1N NaOH	1.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 35–40.0mL volumes.

Use: For the cultivation of *Escherichia coli*.

LB Medium (Luria Broth) (Lenox Broth)

Composition per liter:

Pancreatic digest of casein	10.0g
NaCl	5.0g
Yeast extract	5.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli*.

LB Medium for X1776

Composition per liter:

NaCl	10.0g
Pancreatic digest of casein	10.0g
Yeast extract	5.0g
Glucose solution	10.0mL
Diaminopimelic acid solution	10.0mL
Thymidine solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Glucose Solution:

Composition per 10.0mL:

D-Glucose	0.8g

Preparation of Glucose Solution: Add D-glucose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Diaminopimelic Acid Solution:

Composition per 10.0mL:

DL-Diaminopimelic acid	0.1g

Preparation of Diaminopimelic Acid Solution:

Add diaminopimelic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Thymidine Solution:

Composition per 10.0mL:

Thymidine	0.02g

Preparation of Thymidine Solution:

Add thymidine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except glucose solution, diaminopimelic acid solution, and thymidine solution—to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile glucose solution, diaminopimelic acid solution, and thymidine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Bacillus subtilis* and *Escherichia coli*.

LBE Medium

Composition per liter:

NaCl	10.0g
Pancreatic digest of casein	10.0g
Yeast extract	5.0g
Glucose solution	10.0mL
50X medium E	4.0mL

pH 7.0 ± 0.2 at 25°C

Glucose Solution:

Composition per 100.0mL:

D-Glucose	20.0g

Preparation of Glucose Solution:

Add D-glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

50X Medium E:

Composition per liter:

K ₂ HPO ₄ , anhydrous	500.0g
Na(NH ₄)HPO ₄ ·4H ₂ O	175.0g
Citric acid·H ₂ O	100.0g
MgSO ₄ ·7H ₂ O	10.0g

Preparation of 50X Medium E: Add components to 670.0mL of distilled/deionized water in the following order: MgSO₄·7H₂O, citric acid·H₂O,

K₂HPO₄, and Na(NH₄)HPO₄·4H₂O. Mix thoroughly. Bring volume to 1.0L with distilled/deionized water.

Preparation of Medium: Add components—except glucose solution and 50X medium E—to distilled/deionized water and bring volume to 986.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile glucose solution and 4.0mL of sterile 50X medium E. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Escherichia coli*.

LBS Agar (*Lactobacillus* Selection Agar)

Composition per liter:

Sodium acetate·3H ₂ O.....	25.0g
Glucose	20.0g
Agar	15.0g
Pancreatic digest of casein.....	10.0g
KH ₂ PO ₄	6.0g
Yeast extract.....	5.0g
Ammonium citrate	2.0g
Polysorbate 80.....	1.0g
MgSO ₄	0.575g
FeSO ₄	0.034g
MnSO ₄	0.12g
Acetic acid, glacial.....	1.32mL
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and enumeration of lactobacilli.

LBS Broth (*Lactobacillus* Selection Broth)

Composition per liter:

Sodium acetate·3H ₂ O.....	25.0g
Glucose	20.0g
Pancreatic digest of casein.....	10.0g
KH ₂ PO ₄	6.0g
Yeast extract.....	6.0g
Ammonium citrate	2.0g
Polysorbate 80.....	1.0g
MgSO ₄	0.575g

FeSO ₄	0.034g
MnSO ₄	0.12g
Acetic acid, glacial.....	1.32mL
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Aseptically distribute into sterile tubes.

Use: For the selective isolation and cultivation of lactobacilli.

Lead Acetate Agar

Composition per liter:

Agar	15.0g
Peptone	15.0g
Proteose peptone.....	5.0g
Glucose	1.0g
Lead acetate	0.2g
Na ₂ S ₂ O ₃	0.08g
pH 6.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of Gram-negative coliform bacteria based on H₂S production. Bacteria that produce H₂S turn the medium brown.

Lecithin Lipase Anaerobic Agar

Composition per liter:

Pancreatic digest of casein.....	40.0g
Agar	25.0g
Yeast extract.....	5.0g
Glucose	2.0g
NaCl.....	2.0g
KH ₂ PO ₄	1.0g
Na ₂ HPO ₄ ·12H ₂ O.....	5.0g
MgSO ₄ ·7H ₂ O	0.1g
Egg yolk emulsion	100.0mL
pH 7.6 ± 0.2 at 25°C	

Egg Yolk Emulsion:

Composition:

Chicken egg yolks.....	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Filter sterilize.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and differentiation of *Clostridium* species based on lecithinase production and lipase production. Bacteria that produce lecithinase appear as colonies surrounded by a zone of insoluble precipitate. Bacteria that produce lipase appear as colonies with a pearly iridescent sheen.

Lee's Agar

Composition per liter:

Agar	18.0g
Pancreatic digest of casein	10.0g
Yeast extract	10.0g
Lactose	5.0g
Sucrose	5.0g
CaCO ₃	3.0g
K ₂ HPO ₄	0.5g
Bromcresol Purple (0.2% solution)	10.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Purple Solution:

Composition per 10.0mL:

Bromcresol Purple	0.02g

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except Bromcresol Purple solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 7.0. Gently heat and bring to boiling. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile Bromcresol Purple solution. Mix thoroughly. Pour into sterile, chilled Petri dishes in 20–25mL volumes. Swirl flask while dispensing to evenly suspend CaCO₃. Dry plates at 30°C for 18–24 hr before use.

Use: For the isolation, cultivation, and enumeration of *Lactobacillus bulgaricus* from yogurt.

Leptospira Medium

Composition per liter:

(NH ₄) ₂ Fe(SO ₄) ₂ ·6H ₂ O	6.0g
NaH ₂ PO ₄	0.53g
L-Asparagine	0.5g
Glycerol	0.2g
Tween 60	0.2g
MgSO ₄ ·7H ₂ O	0.15g
KH ₂ PO ₄	0.069g
Tween 80	0.05g
EDTA	0.01g
CaCO ₃	4.0mg
Thiamine·HCl	1.0mg
Vitamin B ₁₂	1.0μg

pH 7.4–7.6 at 25°C

Preparation of Medium: Add components, except thiamine·HCl, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 1.0mg of thiamine·HCl. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leptospira* species.

Leptospira Medium, EMJH

(*Leptospira* Medium,

Ellinghausen-McCullough/
Johnson-Harris)

Composition per liter:

Na ₂ HPO ₄	1.0g
NaCl	1.0g
KH ₂ PO ₄	0.3g
NH ₄ Cl	0.25g
Thiamine	5.0mg
Rabbit serum	100.0mL

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except rabbit serum, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add sterile rabbit serum. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Leptospira* species.

Leptospira Medium, Modified

Composition per liter:

Agar	1.5g
NaCl	0.5g

Peptone.....	0.3g
Beef extract.....	0.2g
Hemin solution.....	2.5mL
Sterile rabbit serum.....	100.0mL
pH 7.3 ± 0.1 at 25°C	

Hemin Solution:**Composition per 100.0mL:**

Hemin.....	0.05g
NaOH (1N solution).....	1.0mL

Preparation of Hemin Solution: Add hemin to 1.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components, except hemin solution and rabbit serum, to distilled/deionized water and bring volume to 897.5mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.4. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2.5mL of sterile hemin solution and 100.0mL of sterile rabbit serum. Mix thoroughly. The pH of the medium should be 7.3. Store at 4°C for 24 hr. Inactivate medium at 56°C for 60 min. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Leptospira biflexa*, *Leptospira borgpetersenii*, *Leptospira interrogans*, *Leptospira meyeri*, *Leptospira noguchii*, *Leptospira santarosai*, and *Leptospira weili*.

***Leptospira* Protein-Free Medium (*Leptospira* PF Medium)**

Composition per liter:

TES (N-Tris[hydroxymethyl]methyl-2-aminoethane sulfonic acid) buffer	1.2g
NaCl.....	0.9g
Sodium pyruvate.....	0.2g
CT-Tween 60.....	12.0mL
CT-Tween 40.....	3.0mL
MgCl ₂ -CaCl ₂ solution.....	1.0mL
Cyanocobalamin (0.02% solution)	1.0mL
Glycerol (10% solution).....	1.0mL
KH ₂ PO ₄ (1% solution).....	1.0mL
MnSO ₄ ·H ₂ O (0.1% solution)	1.0mL
ZnSO ₄ (0.4% solution).....	0.1mL
pH 7.6 ± 0.2 at 25°C	

CT-Tween 60:**Composition per 200.0mL:**

Charcoal, Norit A.....	40.0g
Tween 60.....	20.0g

Preparation of CT-Tween 60: Add Tween 60 to 200.0mL of distilled/deionized water. Mix thorough-

ly. While stirring, add charcoal. Stir mixture for 18 hr at 25°C. Allow charcoal to settle out of suspension for 18 hr at 4°C. Carefully decant the Tween solution off the sediment. Centrifuge the Tween solution at 10,000 × g for 1 hr. Decant supernatant solution. Pass Tween solution through a thin-channel ultrafiltration XM 100 membrane. Store stock solution at –20°C.

CT-Tween 40:**Composition per 200.0mL:**

Charcoal, Norit A.....	40.0g
Tween 40.....	20.0g

Preparation of CT-Tween 40: Add Tween 40 to 200.0mL of distilled/deionized water. Mix thoroughly. While stirring, add charcoal. Stir mixture for 18 hr at 25°C. Allow charcoal to settle out of suspension for 18 hr at 4°C. Carefully decant the Tween solution off the sediment. Centrifuge the Tween solution at 10,000 × g for 1 hr. Decant supernatant solution. Pass Tween solution through a thin-channel ultrafiltration XM 100 membrane. Store stock solution at –20°C.

MgCl₂-CaCl₂ Solution:**Composition per 100.0mL:**

CaCl ₂ ·2H ₂ O	1.5g
MgCl ₂ ·6H ₂ O	1.5g

Preparation of MgCl₂-CaCl₂ Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leptospira* species.

Letheen Agar, Modified (BAM M78)

Composition per liter:

Agar	20.0g
Thiotone	10.0g
Pancreatic digest of casein.....	10.0g
Tween 80.....	7.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Yeast extract.....	2.0g
Glucose	1.0g
Lecithin.....	1.0g
NaHSO ₃	0.1g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For determination of the antimicrobial activity of quaternary ammonium compounds.

Letheen Broth, Modified (BAM M79)

Composition per liter:

Peptic digest of animal tissue.....	10.0g
Thiotone peptone	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g
Tween 80.....	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	2.0g
Lecithin	0.7g
NaHSO ₃	0.1g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped bottles in 90.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For determination of the antimicrobial activity of quaternary ammonium compounds.

***Leuconostoc* Medium**

Composition per liter:

CaCO ₃	50.0g
Malt extract.....	50.0g
Agar	15.0g
NaCl.....	2.5g
Beef extract.....	1.0g
Polypeptone	1.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Leuconostoc mesenteroides*.

***Leuconostoc oenos* Medium**

Composition per liter:

Tryptic digest of casein.....	10.0g
Glucose	10.0g
Fructose.....	5.0g
Yeast extract.....	5.0g
Diammonium citrate	3.5g
L-Cysteine-HCl	0.5g
MgSO ₄ ·7H ₂ O.....	200.0mg

MnSO ₄ ·H ₂ O.....	50.0mg
Tomato juice, filtered.....	100.0mL
Tween 80.....	1.0mL

pH 4.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 4.8. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus oenos*.

***Leuconostoc oenos* Medium**

Composition per liter:

Glucose	10.0g
Peptone	10.0g
Yeast extract.....	5.0g
MnSO ₄ ·4H ₂ O.....	0.1g
Tomato juice	250.0mL
L-Cysteine-HCl solution	10.0mL

pH 4.8 ± 0.2 at 25°C

L-Cysteine-HCl Solution:

Composition per 10.0mL:

L-Cysteine-HCl	0.5g

Preparation of L-Cysteine-HCl Solution: Add L-cysteine-HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteine-HCl solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add sterile L-cysteine-HCl solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Leuconostoc oenos*.

***Leuconostoc oenos* Medium**

Composition per liter:

Glucose	10.0g
Tryptone.....	10.0g
Fructose.....	5.0g
Yeast extract.....	5.0g
Diammonium citrate	3.5g
L-Cysteine-HCl	0.5g
So ₄ Mg·7H ₂ O.....	0.2g
MnSO ₄ ·H ₂ O.....	0.05g
Tomato juice, filtered.....	100.0mL
Tween 80.....	1.0mL

pH 4.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 4.8. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Leuconostoc oenos*.

Levine EMB Agar
(Levine Eosin Methylene Blue Agar)
(Eosin Methylene Blue Agar, Levine)
(LEMB Agar)
(BAM M80)

Composition per liter:

Agar	15.0g
Lactose	10.0g
Peptone.....	10.0g
K ₂ HPO ₄	2.0g
Eosin Y	0.4g
Methylene Blue.....	0.065mg

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and differentiation of Gram-negative enteric bacteria based on lactose fermentation. Bacteria that ferment lactose, especially the coliform bacterium *Escherichia coli*, appear as colonies with a green metallic sheen or blue-black to brown color. Bacteria that do not ferment lactose appear as colorless or transparent light purple colonies.

LICNR Broth

(Lysine Iron Cystine Neutral Red Broth)

Composition per 500.0mL:

L-Lysine·HCl	10.0g
Mannitol.....	5.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	3.0g
Glucose	1.0g
Salicin	1.0g
Ferric ammonium citrate.....	0.5g
L-Cystine	0.1g
Na ₂ S ₂ O ₃	0.1g
Neutral Red	0.025g
Novobiocin solution.....	10.0mL

pH 6.2 ± 0.2 at 25°C

Novobiocin Solution:

Composition per 10.0mL:

Novobiocin.....	0.015g

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.1mL of sterile novobiocin solution to each tube. Mix thoroughly.

Use: For the rapid, presumptive detection of *Salmonella* in foods, food ingredients, and feed materials.

Listeria Enrichment Broth

Composition per liter:

Pancreatic digest of casein.....	17.0g
Yeast extract.....	6.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
Glucose	2.5g
K ₂ HPO ₄	2.5g
Cycloheximide.....	0.05g
Nalidixic acid.....	0.04g
Acriflavine·HCl	0.015g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Listeria monocytogenes* according to the FDA formula.

Listeria Enrichment Broth, FDA
(LEB, FDA)

Composition per liter:

Soybean casein digest broth yeast extract	1.0L
Nalidixic acid solution	8.0mL
Cycloheximide solution.....	5.1mL
Acriflavin·HCl solution	3.0mL

pH 7.3 ± 0.2 at 25°C

Soybean Casein Digest Broth Yeast Extract:

Composition per liter:

Pancreatic digest of casein.....	17.0g
Yeast extract.....	6.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Soybean Casein Digest Broth

Yeast Extract: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Nalidixic Acid Solution:

Composition per 100.0mL:

Nalidixic acid.....0.5g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 100.0mL:

Cycloheximide.....1.5g

Ethanol.....40.0mL

Preparation of Cycloheximide Solution: Add cycloheximide to 40.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Acriflavin-HCl Solution:

Composition per 100.0mL:

Acriflavin-HCl solution.....0.5g

Preparation of Acriflavin-HCl Solution: Add acriflavin-HCl solution to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except nalidixic acid solution, acriflavin solution, and cycloheximide solution—to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 8.0mL of sterile nalidixic acid solution, 5.1mL of sterile cycloheximide solution, and 3.0mL of sterile acriflavin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and enrichment of *Listeria monocytogenes* from foods.

Listeria Enrichment Broth I, USDA FSIS

(Listeria Primary Selective

Enrichment Broth, UVM I)

(University of Vermont I Listeria

Primary Selective Enrichment Broth)

Composition per liter:

NaCl.....20.0g

Na₂HPO₄.....12.0g

Beef extract.....5.0g

Protease peptone.....5.0g

Pancreatic digest of casein.....5.0g

Yeast extract.....5.0g

KH₂PO₄.....1.35g

Esculin.....1.0g

Nalidixic acid solution.....1.0mL

Acriflavine solution.....1.0mL

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid.....0.2g

NaOH (0.1M solution).....10.0mL

Preparation of Nalidixic Acid Solution: Add nalidixic acid to 10.0mL of NaOH solution. Mix thoroughly. Filter sterilize.

Acriflavine Solution:

Composition per 10.0mL:

Acriflavine.....0.12g

Preparation of Acriflavine Solution: Add acriflavine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: Add components, except nalidixic acid solution and acriflavine solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile nalidixic acid solution. Mix thoroughly. Store at 4°C. Immediately prior to use, aseptically add 1.0mL of sterile acriflavine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the selective isolation, cultivation, and enrichment of *Listeria monocytogenes* from food, milk, and dairy products.

Listeria Enrichment Broth II,

USDA FSIS

(Listeria Primary Selective

Enrichment Broth, UVM II)

(University of Vermont II Listeria

Primary Selective Enrichment Broth

Composition per liter:

NaCl.....20.0g

Na₂HPO₄.....12.0g

Beef extract.....5.0g

Protease peptone.....5.0g

Pancreatic digest of casein.....5.0g

Yeast extract.....5.0g

KH₂PO₄.....1.35g

Esculin.....1.0g

Nalidixic acid solution	1.0mL
Acriflavine solution	1.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid.....	0.2g
NaOH (0.1M solution)	10.0mL

Preparation of Nalidixic Acid Solution: Add nalidixic acid to 10.0mL of NaOH solution. Mix thoroughly. Filter sterilize.

Acriflavine Solution:

Composition per 10.0mL:

Acriflavine	0.25g

Preparation of Acriflavine Solution: Add acriflavine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Preparation of Medium: Add components, except nalidixic acid solution and acriflavine solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to $45^{\circ}\text{--}50^{\circ}\text{C}$. Aseptically add sterile nalidixic acid solution. Mix thoroughly. Store at 4°C . Immediately prior to use, aseptically add 1.0mL of sterile acriflavine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the selective isolation, cultivation, and enrichment of *Listeria monocytogenes* from food, milk and dairy products.

***Listeria* Fermentation Broth**

Composition per liter:

Proteose peptone No. 3	10.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Bromcresol Purple	0.1g
Carbohydrate solution.....	10.0mL

Carbohydrate Solution:

Composition per 10.0mL:

Carbohydrate.....	5.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Use glucose, salicin, rhamnose, dulcitol, or raffinose.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15

min at 15 psi pressure– 121°C . Cool to $45^{\circ}\text{--}50^{\circ}\text{C}$. Aseptically add sterile carbohydrate solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and differentiation of *Listeria* species based on the fermentation of glucose, salicin, rhamnose, dulcitol, and raffinose.

***Listeria* Transport Enrichment Medium**

Composition per liter:

Sodium glycerophosphate.....	10.0g
Agar	2.0g
Sodium thioglycolate	1.0g
CaCl_2	0.1g
Nalidixic acid.....	0.04g
Acridine solution	2.0mL
pH 7.4 ± 0.2 at 25°C	

Acridine Solution:

Composition per 10.0mL:

Acridine	0.04g

Preparation of Acridine Solution: Add acridine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to $45^{\circ}\text{--}50^{\circ}\text{C}$.

Preparation of Medium: Add components, except acridine solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to $45^{\circ}\text{--}50^{\circ}\text{C}$. Aseptically add acridine solution. Mix thoroughly. Aseptically distribute into sterile tubes in 10.0mL volumes or fill bottles 4/5's full.

Use: For the maintenance—as a transport medium—and enrichment of *Listeria* species.

Litmus Lactose Agar (LL Agar)

Composition per liter:

Agar	10.0g
Lactose.....	10.0g
Meat peptone	5.0g
Beef extract.....	3.0g
Litmus	1.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the maintenance of lactic acid bacteria and differentiation of bacteria on the basis of lactose fermentation. Bacteria that ferment lactose appear as red colonies and others as dark blue-purple colonies.

Litmus Lactose Agar with Crystal Violet (LLK Agar)

Composition per liter:

Agar	10.0g
Lactose	10.0g
Meat peptone.....	5.0g
Beef extract.....	3.0g
Litmus	1.0g
Crystal Violet.....	5.0mg
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the maintenance of lactic acid bacteria and for the differentiation of several bacteria on the basis of lactose fermentation. Bacteria that ferment lactose appear as red colonies. Bacteria that do not ferment lactose appear as dark blue-purple colonies.

Litmus Milk

Composition per liter:

Skim milk.....	100.0g
Azolitmin	0.5g
Na ₂ SO ₃	0.5g
pH 6.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 20 min at 10 psi pressure–115°C.

Use: For the maintenance of lactic acid bacteria and for the differentiation of several bacteria, especially *Clostridium* species, based on their action on milk. Bacteria that do not ferment carbohydrates, such as *Proteus vulgaris* or *Moraxella lacunata*, show no change in the litmus indicator. Bacteria that ferment lactose or glucose with the production of gas, such as *Clostridium perfringens*, turn the medium pink and frothy. Bacteria that proteolytically degrade lactalbumin turn the medium blue. Bacteria that coagulate casein form a curd or clot. Bacteria that peptonize

casein, such as *Pseudomonas aeruginosa*, show a dissolution of the clot.

Liver Broth

Composition per liter:

Extracted liver tissue, minced.....	30.0g
Infusion from fresh liver.....	23.0g
Peptone	10.0g
K ₂ HPO ₄	1.0g
Agar overlay solution	200.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Agar Overlay Solution:

Composition per 200.0mL:

Agar, Oxoid No. 3.....	4.0g

Preparation of Agar Overlay Solution: Add agar to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components, except agar, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes to a depth of 50mm. Make sure that some liver particles are transferred to each tube. Autoclave for 20 min at 10 psi pressure–115°C. After inoculation, aseptically overlayer the broth with 2.0mL of sterile, cooled agar solution per tube.

Use: For the isolation and cultivation of saccharolytic or putrefactive mesophilic and thermophilic anaerobic bacteria from foods.

Liver Broth

Composition per liter:

Beef liver, fresh.....	453.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	1.0g
Soluble starch.....	1.0g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Remove the fat from fresh beef liver. Grind the liver. Add 1.0L of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 60 min. Adjust pH to 7.6. Filter through cheesecloth. Reserve meat. To filtrate, add pancreatic digest of casein, K₂HPO₄, and soluble starch. Bring volume to 1.0L with distilled/deionized water. Refilter solution. Add meat particles to test tubes to a depth of approximately 2cm. Distribute broth into tubes with meat particles in 15.0mL volumes. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of anaerobic microorganisms, especially *Clostridium botulinum*, from foods.

Liver Broth with NaCl

Composition per liter:

Beef liver, fresh.....	453.0g
NaCl.....	150.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	1.0g
Soluble starch.....	1.0g

pH 7.6 ± 0.2 at 25°C

Preparation of Medium: Remove the fat from fresh beef liver. Grind the liver. Add 1.0L of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 60 min. Adjust pH to 7.6. Filter through cheesecloth. Reserve meat. To filtrate, add pancreatic digest of casein, K₂HPO₄, NaCl, and soluble starch. Bring volume to 1.0L with distilled/deionized water. Refilter solution. Add meat particles to test tubes to a depth of approximately 2cm. Distribute broth into tubes with meat particles in 15.0mL volumes. Autoclave for 20 min at 15 psi pressure–121°C. After inoculation, overlay each tube with sterile, melted petroleum jelly.

Use: For the cultivation of obligate halophiles from brined and dry-salted vegetables.

Liver Infusion Sake Medium

Composition per liter:

Liver, fresh.....	400.0g
Agar.....	15.0g
Sake.....	400.0mL

Preparation of Medium: Finely mince liver and add to 600.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 5–10 min. Filter through Whatman #2 filter paper. To filtrate, add agar and sake. Bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus fructivorans* and *Lactobacillus homohiochi*.

Liver Veal Agar (BAM M83)

Composition per liter:

Veal, infusion from.....	500.0g
Beef liver, infusion from.....	50.0g
Gelatin.....	20.0g

Proteose peptone.....	20.0g
Agar.....	15.0g
Soluble starch.....	10.0g
Glucose.....	5.0g
NaCl.....	5.0g
Casein.....	2.0g
NaNO ₃	2.0g
Enzymatic digest of protein.....	1.3g
Pancreatic digest of casein.....	1.3g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Make sure that some liver and veal particles are transferred to each tube. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of anaerobic organisms.

Liver Veal Egg Yolk Agar (BAM M84)

Composition per 1080.0mL:

Liver veal agar.....	1.0L
Egg yolk emulsion, 50%.....	80.0mL

pH 7.3 ± 0.2 at 25°C

Liver Veal Agar:

Composition per liter:

Veal, infusion from.....	500.0g
Beef liver, infusion from.....	50.0g
Gelatin.....	20.0g
Proteose peptone.....	20.0g
Agar.....	15.0g
Soluble starch.....	10.0g
Glucose.....	5.0g
NaCl.....	5.0g
Casein.....	2.0g
NaNO ₃	2.0g
Enzymatic digest of protein.....	1.3g
Pancreatic digest of casein.....	1.3g

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Liver Veal Agar: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Make sure that some liver and veal particles are transferred to each tube. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg.....	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: To 1.0L of cooled sterile liver veal agar, aseptically add 80.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes. Dry plates at 35°C for 24 hr.

Use: For the cultivation of a variety of anaerobic organisms.

LMX Broth Modified, Fluorocult (Fluorocult LMX Broth, Modified)

Composition per liter:

Tryptose	5.0
NaCl	5.0
K ₂ HPO ₄	2.7g
KH ₂ PO ₄	2.0g
Sorbitol.....	1.0
Tryptophan.....	1.0
Lauryl sulfate sodium salt.....	0.1
5-Bromo-4-chloro-3-indolyl-β-D-galactopyranoside (X-GAL)	0.08
4-Methylumbelliferyl-β-D-glucuronide	0.05g
1-Isopropyl-β-D-1-thio-galactopyranoside (IPTG)	0.1

pH: 6.8 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes. Autoclave for 15 min at 15 psi pressure–121°C. The broth is clear and yellowish brown.

Use: For the simultaneous detection of total coliforms and *E. coli* in water, food and dairy products by the fluorogenic procedure. A color change of the broth from yellow to blue-green indicates the presence of coliforms. A blue fluorescence under long-wave UV light permits the rapid detection of *E. coli*. As tryptophan is added to the broth, the indole reaction is easily done by adding KOVACS reagent. The formation of a red ring additionally confirms the presence of *E. coli*.

Long-Term Preservation Medium (BAM M85)

Composition per liter:

NaCl.....	30.0g
Peptone	10.0g
Agar	3.0g
Yeast extract.....	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a wide variety of bacteria.

Lowenstein-Gruft Medium

Composition per 1600.0mL:

Potato starch.....	30.0g
Asparagine	3.6g
KH ₂ PO ₄	2.4g
Magnesium citrate	0.6g
Malachite Green.....	0.4g
MgSO ₄ ·7H ₂ O	0.24g
Nalidixic acid.....	0.056g
Ribonucleic acid	0.08mg
Homogenized whole egg	1.0L
Glycerol	12.0mL
Penicillin	80,000U

Homogenized Whole Egg:**Composition per liter:**

Whole eggs	18–24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except fresh egg mixture. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 1.0L of homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation and differentiation of *Mycobacterium* species. *Mycobacterium tuberculosis* ap-

pears as granular, rough, dry colonies. *Mycobacterium kansasii* appears as smooth to rough photochromogenic colonies. *Mycobacterium gordonae* appears as smooth yellow-orange colonies. *Mycobacterium avium* appears as smooth, colorless colonies. *Mycobacterium smegmatis* appears as wrinkled, creamy white colonies.

Lowenstein-Jensen Medium

Composition per 1600.0mL:

Potato starch.....	30.0g
Asparagine	3.6g
KH ₂ PO ₄	2.4g
Magnesium citrate.....	0.6g
Malachite Green.....	0.4g
MgSO ₄ ·7H ₂ O	0.24g
Homogenized whole egg	1.0L
Glycerol	12.0mL

Source: This medium is available as a prepared medium from BD Diagnostic Systems and Oxoid.

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18-24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except fresh egg mixture. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 1.0L of homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation and differentiation of *Mycobacterium* species. *Mycobacterium tuberculosis* appears as granular, rough, dry colonies. *Mycobacterium kansasii* appears as smooth to rough photochromogenic colonies. *Mycobacterium gordonae* appears as smooth yellow-orange colonies. *Mycobacterium avium* appears as smooth, colorless colonies. *Mycobacterium smegmatis* appears as wrinkled, creamy white colonies. Also used for the cultivation and maintenance of *Gordona* species, *Nocardia* species, *Rhodococcus* species, and *Tsukamurella paurometabolum*.

Lowenstein-Jensen Medium with NaCl

Composition per 1600.0mL:

NaCl.....	80.0g
Potato starch.....	30.0g
Asparagine	3.6g
KH ₂ PO ₄	2.4g
Magnesium citrate	0.6g
Malachite Green.....	0.4g
MgSO ₄ ·7H ₂ O	0.24g
Homogenized whole egg	1.0L
Glycerol	12.0mL

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18-24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium:

Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except fresh egg mixture. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 1.0L of homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation of *Mycobacterium smegmatis* and other salt-tolerant *Mycobacterium* species.

Lowenstein-Jensen Medium with Streptomycin

Composition per 161.0mL:

Potato starch.....	30.0g
Asparagine	3.6g
KH ₂ PO ₄	2.4g
Magnesium citrate	0.6g
Malachite Green.....	0.4g
MgSO ₄ ·7H ₂ O	0.24g
Homogenized whole egg	1.0L
Glycerol	12.0mL
Streptomycin solution.....	10.0mL

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18-24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min.

Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Streptomycin Solution:

Composition per 10.0mL:

Streptomycin 0.1mg

Preparation of Streptomycin Solution: Add streptomycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 600.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except fresh egg mixture. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 1.0L of homogenized whole egg and 10.0mL of sterile streptomycin solution. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation and differentiation of *Mycobacterium* species. *Mycobacterium tuberculosis* appears as granular, rough, dry colonies. *Mycobacterium kansasii* appears as smooth to rough photochromogenic colonies. *Mycobacterium gordonae* appears as smooth yellow-orange colonies. *Mycobacterium avium* appears as smooth, colorless colonies. *Mycobacterium smegmatis* appears as wrinkled, creamy white colonies. Also used for the cultivation and maintenance of *Gordona* species, *Nocardia* species, *Rhodococcus* species, and *Tsukamurella paurometabolum*.

Lowenstein-Jensen Medium without Glycerol

Composition per liter:

Potato starch 30.0g
Asparagine 3.6g
KH₂PO₄ 2.4g
Magnesium citrate 0.6g
Malachite green 0.4g
MgSO₄·7H₂O 0.24g
Homogenized whole egg 1.0L

Homogenized Whole Egg:

Composition per liter:

Whole eggs 18–24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Ho-

mogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add components, except fresh egg mixture, to 600.0mL of distilled/deionized water. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 1.0L of homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation and maintenance of *Mycobacterium* species, especially *Mycobacterium bovis* and other species that are sensitive to glycerol.

LPM Agar (Lithium Chloride Phenylethanol Moxalactam Plating Agar) (BAM M81)

Composition per liter:

Agar 15.0g
Glycine anhydride 10.0g
LiCl 5.0g
NaCl 5.0g
Pancreatic digest of casein 5.0g
Peptic digest of animal tissue 5.0g
Beef extract 3.0g
Phenylethyl alcohol 2.5g
Moxalactam solution 2.0mL
pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Moxalactam Solution:

Composition per 10.0mL:

Moxalactam 0.1g

Preparation of Moxalactam Solution: Add moxalactam to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except moxalactam solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 12 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2.0mL of sterile moxalactam solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes*.

LPM Agar with Esculin and Ferric Iron (BAM M82)

Composition per liter:

Agar	15.0g
Glycine anhydride	10.0g
LiCl	5.0g
NaCl	5.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	3.0g
Phenylethyl alcohol	2.5g
Esculin	1.0g
Ferric ammonium citrate	0.5g
Moxalactam solution	2.0mL
pH 7.3 ± 0.2 at 25°C	

Moxalactam Solution:

Composition per 10.0mL:

Moxalactam	0.1g

Preparation of Moxalactam Solution: Add moxalactam to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except moxalactam solution, to distilled/deionized water and bring volume to 998.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 12 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 2.0mL of sterile moxalactam solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes*.

LS Differential Medium (*Lactobacillus Streptococcus* Differential Medium)

Composition per liter:

Glucose	20.0g
Agar	13.0g
Pancreatic digest of casein	10.0g
Beef extract	5.0g
NaCl	5.0g
Papaic digest of soybean meal	5.0g
Yeast extract	5.0g
L-Cysteine·HCl·H ₂ O	0.3g
Skim milk solution	100.0mL
Triphenyltetrazolium chloride solution	10.0mL
pH 6.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Skim Milk Solution:

Composition per 100.0mL:

Skim milk, antibiotic free	10.0g

Preparation of Skim Milk Solution: Add skim milk to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 5 min at 15 psi pressure–121°C. Cool to 50°C.

Triphenyltetrazolium Chloride Solution:

Composition per 10.0mL:

2,3,5-Triphenyltetrazolium chloride	0.2g

Preparation of Triphenyltetrazolium Chloride Solution: Add triphenyltetrazolium chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except skim milk solution and triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile skim milk solution and sterile triphenyltetrazolium chloride solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the differentiation and enumeration of lactobacilli and streptococci in yogurt. *Lactobacillus* species appear as irregular, red colonies surrounded by a white, opaque zone. *Streptococcus* species appear as round, red colonies surrounded by a clear zone.

Lysine Agar, Selective

Composition per liter:

Agar	15.0g
L-Lysine	10.0g
Peptone	5.0g
Glucose	3.5g
Yeast extract	3.0g
Bile salts mixture	1.5g
Sulfapyridine	0.3g
Bromocresol Purple	0.03g
Crystal Violet	0.001g
pH 6.8 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation and cultivation of *Salmonella* species from food by the hydrophobic grid membrane filter method.

**Lysine Arginine Iron Agar
(LAIA)
(BAM M86)**

Composition per liter:

Agar	15.0g
L-Arginine	10.0g
L-Lysine	10.0g
Peptone	5.0g
Yeast extract	3.0g
Glucose	1.0g
Ferric ammonium citrate	0.5g
Sodium thiosulfate	0.04g
Bromocresol Purple	0.02g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.8. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave for 12 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate lysine, decarboxylate arginine, and produce H₂S. Bacteria that decarboxylate lysine or arginine turn the medium purple. Bacteria that produce H₂S appear as black colonies.

**Lysine Decarboxylase Broth, Falkow
(BAM M44)/(BAM M87)**

Composition per liter:

Peptone	5.0g
L-Lysine	5.0g
Yeast extract	3.0g
Glucose	1.0g
Bromocresol Purple	0.02g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.5–6.8. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria, especially *Salmonella*, based on their ability to decarboxylate lysine. Bacteria that decarboxylate lysine turn the medium turbid purple.

**Lysine Broth Falkow with NaCl
(BAM M44)/(BAM M87)**

Composition per liter:

L-Lysine	5.0g
Peptone or gelysate	5.0g
Yeast extract	3.0g
Glucose	1.0g
Bromocresol Purple	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure–121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of *Vibrio* spp. based on their ability to decarboxylate the amino acid lysine. Bacteria that decarboxylate lysine turn the medium turbid purple.

**Lysine Decarboxylase Broth,
Taylor Modification**

Composition per liter:

L-Lysine	5.0g
Yeast extract	3.0g
Glucose	1.0g
Bromocresol Purple	0.02g
pH 6.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.1. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria, especially *Salmonella*, based on their ability to decarboxylate lysine. Bacteria that decarboxylate lysine turn the medium turbid purple.

**Lysine Decarboxylase Broth,
Taylor Modification
(Lysine Decarboxylase Broth)**

Composition per liter:

L-Lysine	5.0g
Peptone	5.0g
Yeast extract	3.0g

Glucose	1.0g
Bromocresol Purple	0.02g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.1. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure— 121°C .

Use: For the cultivation and differentiation of bacteria, especially *Salmonella*, based on their ability to decarboxylate lysine. Bacteria that decarboxylate lysine turn the medium turbid purple.

Lysine Decarboxylase Medium (LDC Medium) (BAM M88)

Composition per liter:

Glucose	0.5g
KH_2PO_4	0.5g
L-Lysine-HCl	0.5g
pH 4.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 4.6. Autoclave for 15 min at 15 psi pressure— 121°C . Aseptically distribute into sterile tubes in 1.0mL volumes.

Use: For the cultivation and differentiation of Gram-negative, nonfermentative bacteria based on their ability to decarboxylate lysine. Bacteria that decarboxylate lysine turn the medium turbid purple.

Lysine Iron Agar, Edward and Fife (BAM M89)

Composition per liter:

Agar	13.5g
L-Lysine	10.0g
Pancreatic digest of gelatin	5.0g
Yeast extract	3.0g
Glucose	1.0g
Ferric ammonium citrate	0.5g
$\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$	0.04g
Bromocresol Purple	0.02g
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 12 min at 15 psi pressure— 121°C . Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of members of the Enterobacteriaceae based on their ability to decarboxylate lysine and to form H_2S . Bacteria that decarboxylate lysine turn the medium purple. Bacteria that produce H_2S appear as black colonies.

Lysine Medium

Composition per liter:

Glucose	44.5g
Agar	17.8g
KH_2PO_4	1.78g
Lysine	1.0g
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	0.89g
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	0.178g
NaCl	0.089g
Inositol	0.02g
Calcium pantothenate	2.0mg
Adenine	1.78mg
DL-Methionine	0.89mg
L-Histidine	0.89mg
DL-Tryptophan	0.89mg
Thiamine-HCl	0.4mg
Pyridoxine	0.4mg
Nicotinic acid	0.4mg
$\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$	0.22mg
p-Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	0.035mg
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.035mg
$(\text{NH}_4)_2\text{MoO}_4 \cdot 4\text{H}_2\text{O}$	0.018mg
H_3BO_3	8.9 μg
Biotin	2.0 μg
Folic acid	1.0 μg
Potassium lactate (50% solution)	10.0mL
Lactic acid	1.0mL

pH 4.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add potassium lactate to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Add remaining components, except lactic acid. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 50°C . Adjust pH to 4.8 by adding 1.0mL of lactic acid. Pour into sterile Petri dishes. Dry the surface of the plates by incubation at 37°C for 24 hr.

Use: For the isolation, cultivation, and enumeration of wild yeasts encountered in brewing. For the isolation and enumeration of wild yeasts encountered in

brewing. On this medium, pitching yeasts are suppressed.

Lysine Ornithine Mannitol Agar (LOM Agar)

Composition per liter:

Agar	13.5g
L-Ornithine-HCl	6.5g
D-Mannitol	5.25g
L-Lysine-HCl	5.0g
NaCl	5.0g
Yeast extract	3.0g
Bromthymol Blue	0.3g
Vancomycin solution	10.0mL
pH 6.5 ± 0.2 at 25°C	

Vancomycin Solution:

Composition per 10.0mL:

Vancomycin-HCl	0.03g

Preparation of Vancomycin Solution: Add vancomycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except vancomycin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile vancomycin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and differentiation of Gram-negative bacilli based on their ability to decarboxylate lysine or ornithine and mannitol fermentation. Especially useful for the identification of *Enterobacter agglomerans*. Bacteria that ferment mannitol appear as dark yellow colonies. Bacteria that decarboxylate lysine or ornithine appear as green-yellow colonies.

Lysozyme Broth (BAM M90)

Composition per 1010.0mL:

Nutrient broth	1.0L
Lysozyme solution	10.0mL
pH 6.9 ± 0.2 at 25°C	

Nutrient Broth:

Composition per liter:

Pancreatic digest of gelatin	5.0g
Beef extract	3.0g

Source: Nutrient broth is available as a premixed powder from BD Diagnostic Systems.

Preparation of Nutrient Broth: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into bottles in 99.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Lysozyme Solution:

Composition per 100.0mL:

Lysozyme	0.1g

Preparation of Lysozyme Solution: Add lysozyme to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add 1.0mL of sterile lysozyme solution to 99.0mL of cooled, sterile nutrient broth. Mix thoroughly. Aseptically distribute into sterile tubes in 2.5mL volumes.

Use: For the cultivation and differentiation of *Bacillus cereus* in foods. *Bacillus cereus* is resistant to lysozyme and will grow in this medium.

M16 Agar

Composition per liter:

Agar	10.0g
Beef extract	5.0g
Pancreatic digest of soybean meal	5.0g
Polypeptone	5.0g
Sodium acetate·3H ₂ O	3.0g
Yeast extract	2.5g
Ascorbic acid	0.5g
Carbohydrate solution	50.0mL
pH 7.2 ± 0.2 at 25°C	

Carbohydrate Solution:

Composition per 50.0mL:

Carbohydrate	5.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 50.0mL. Lactose or glucose may be used. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.2 with 2N NaOH. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of lactobacilli from cheddar cheese.

M17 Agar (LMG Medium 261)

Composition per liter:

Disodium β -glycerophosphate	19.0g
Agar	11.0g
Polypeptone	5.0g
Beef extract	5.0g
Papaic digest of soybean meal	5.0g
Yeast extract	2.5g
Ascorbic acid	0.5g
Lactose solution	50.0mL
MgSO ₄ ·7H ₂ O (1M solution)	1.0mL
pH 6.9 $\pm$ 0.2 at 25°C	

Lactose Solution:

Composition per 100.0mL:

Lactose	10.0g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except lactose solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile lactose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Streptococcus thermophilus* and for the cultivation and maintenance of streptococci and their bacteriophages. Also used for the cultivation and maintenance of starter cultures for cheese and yogurt manufacture as well as detecting streptococcal mutants that are unable to ferment lactose.

M17 Agar

Composition per liter:

Disodium β -glycerophosphate	19.0g
Agar	11.0g
Beef extract	5.0g
Papaic digest of soybean meal	5.0g
Yeast extract	2.5g
Ascorbic acid	0.5g
MgSO ₄ ·7H ₂ O	0.25g
Lactose solution	50.0mL
pH 6.9 $\pm$ 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Lactose Solution:

Composition per 100.0mL:

Lactose	10.0g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except lactose solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile lactose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of streptococci and their bacteriophages. Also used for the cultivation and maintenance of starter cultures for cheese and yogurt manufacture as well as detecting streptococcal mutants which are unable to ferment lactose. For improved growth of lactic streptococci and their bacteriophages and selective enumeration of *Streptococcus thermophilus* from yogurt.

M56 Agar

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	8.7g
KH ₂ PO ₄	5.3g
D-Glucose	4.0g
(NH ₄) ₂ SO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.1g
L-Histidine	0.05g
L-Leucine	0.05g
Uracil	0.03g
Ca(NO ₃) ₂ ·4H ₂ O	5.0mg
FeSO ₄ ·7H ₂ O	5.0mg
ZnSO ₄ ·7H ₂ O	5.0mg
pH 7.0 $\pm$ 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

M Broth

Composition per liter:

Pancreatic digest of casein	12.5g
K ₂ HPO ₄	5.0g
NaCl	5.0g
Sodium citrate	5.0g
Yeast extract	5.0g
Mannose	2.0g
MgSO ₄ ·7H ₂ O	0.8g

Polysorbate 80.....	0.75g
FeSO ₄	0.04g
pH 7.0 ± 0.22 at 25°C	

Source: Available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the detection of *Salmonella* in dried foods and feeds.

M10 Broth

Composition per liter:

Pancreatic digest of casein.....	2.0g
Yeast extract.....	2.0g
Cellobiose.....	1.0g
Glucose.....	1.0g
Maltose.....	1.0g
Starch.....	1.0g
Resazurin.....	1.0mg
Mineral solution I.....	100.0mL
Mineral solution II.....	100.0mL
Na ₂ CO ₃ solution.....	50.0mL
Hemin solution.....	10.0mL
L-Cysteine·HCl solution.....	10.0mL
Volatile fatty acid mixture.....	3.1mL
pH 6.8 ± 0.2 at 25°C	

Mineral Solution I:

Composition per 100.0mL:

K ₂ HPO ₄	0.2g

Preparation of Mineral Solution I: Add K₂HPO₄ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Mineral Solution II:

Composition per 100.0mL:

NaCl.....	0.4g
(NH ₄) ₂ SO ₄	0.4g
KH ₂ PO ₄	0.3g
MgSO ₄ ·7H ₂ O.....	0.09g
CaCl ₂	0.05g

Preparation of Mineral Solution II: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Na₂CO₃ Solution:

Composition per 100.0mL:

Na ₂ CO ₃	8.0g

Preparation of Na₂CO₃ Solution: Add Na₂CO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Sparge with 100% CO₂. Autoclave for 15 min at 15 psi pressure–121°C.

Hemin Solution:

Composition per 100.0mL:

Hemin.....	1.0g
NaOH (1N solution).....	10.0mL

Preparation of Hemin Solution: Add components to 100.0mL of distilled/deionized water. Mix thoroughly.

L-Cysteine·HCl Solution:

Composition per 10.0mL:

L-Cysteine·HCl.....	0.25g

Preparation of L-Cysteine·HCl Solution: Add L-cysteine·HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave under 100% N₂ for 15 min at 15 psi pressure–121°C.

Volatile Fatty Acid Mixture:

Composition per 31.0mL:

Acetic acid.....	17.0mL
Propionic acid.....	6.0mL
Butyric acid.....	4.0mL
DL-α-Methylbutyric acid.....	1.0mL
Isobutyric acid.....	1.0mL
Isovaleric acid.....	1.0mL
n-Valeric acid.....	1.0mL

Preparation of Volatile Fatty Acid Mixture:

Combine components. Mix thoroughly. Store under 100% N₂.

Preparation of Medium: Prepare and dispense medium under 100% CO₂. Add components, except L-cysteine·HCl solution and Na₂CO₃ solution, to distilled/deionized water and bring volume to 930.0mL. Mix thoroughly. Sparge with 100% CO₂. Adjust pH to 6.8 with 1N NaOH. Distribute anaerobically 9.3mL volumes into Hungate tubes. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically and anaerobically add 0.2mL of sterile L-cysteine·HCl solution and 0.5mL of sterile Na₂CO₃ solution. Check that final pH is 6.8. (Note: if not properly gassed with 100% CO₂, the medium pH can be as high as 9.5.)

Use: For the cultivation of *Enterococcus* species, *Lactobacillus* species and *Streptococcus* species.

M17 Broth

Composition per liter:

Disodium β-glycerophosphate.....	19.0g
Beef extract.....	5.0g
Lactose.....	5.0g
Papaic digest of soybean meal.....	5.0g
Pancreatic digest of casein.....	2.5g
Peptic digest of animal tissue.....	2.5g
Yeast extract.....	2.5g

Ascorbic acid	0.5g
MgSO ₄ ·7H ₂ O	0.25g
pH 7.15 ± 0.05 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of streptococci and their bacteriophages. Also used for the cultivation and maintenance of starter cultures for cheese and yogurt manufacture as well as detecting streptococcal mutants that are unable to ferment lactose.

M9 Medium

Composition per liter:

Na ₂ HPO ₄	6.0g
KH ₂ PO ₄	3.0g
NH ₄ Cl	1.0g
NaCl	0.5g
Glucose solution	10.0mL
MgSO ₄ ·7H ₂ O solution	1.0mL
Thiamine-HCl solution	1.0mL
CaCl ₂ solution	1.0mL
pH 7.0 ± 0.2 at 25°C	

Glucose Solution:

Composition per 100.0mL:

D-Glucose	20.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

MgSO₄·7H₂O Solution:

Composition per liter:

MgSO ₄ ·7H ₂ O	246.5g

Preparation of MgSO₄·7H₂O Solution: Add MgSO₄·7H₂O to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Thiamine-HCl Solution:

Composition per 10.0mL:

Thiamine-HCl	10.0mg

Preparation of Thiamine-HCl Solution: Add thiamine-HCl to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

CaCl₂ Solution:

Composition per liter:

CaCl ₂ solution	14.7g

Preparation of CaCl₂ Solution: Add CaCl₂ solution to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except MgSO₄·7H₂O solution, glucose solution, thiamine-HCl solution, and CaCl₂ solution, to distilled/deionized water and bring volume to 987.0mL. Mix thoroughly. Adjust pH to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature. Aseptically add sterile MgSO₄·7H₂O solution, sterile glucose solution, sterile thiamine-HCl solution, and sterile CaCl₂ solution. Mix thoroughly. Distribute into tubes or flasks.

Use: For the cultivation and maintenance of *Escherichia coli* and a variety of other bacteria.

M56 Medium

Composition per liter:

Na ₂ HPO ₄	8.7g
KH ₂ PO ₄	5.3g
D-Glucose	4.0g
(NH ₄) ₂ SO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.1g
L-Histidine	0.05g
L-Leucine	0.05g
Uracil	0.03g
Ca(NO ₃) ₂ ·4H ₂ O	5.0mg
FeSO ₄ ·7H ₂ O	5.0mg
ZnSO ₄ ·7H ₂ O	5.0mg
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli*.

M17 Medium for Lactic Streptococci (DSMZ Medium 449)

Composition per liter:

Na ₂ -β-glycerophosphate	19.0g
Peptone from casein	5.0g
Soya peptone	5.0g
Peptone bacteriological	5.0g
Yeast extract	2.5g
Ascorbic acid	0.5g
MgSO ₄ ·7H ₂ O	0.25g
Lactose solution	10.0mL
pH 6.9 ± 0.2 at 25°C	

Lactose Solution:

Composition per 10.0mL:

Lactose	5.0g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except lactose solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 10.0mL sterile lactose solution. Aseptically distribute to sterile tubes or flasks.

Use: For the cultivation and maintenance of *Lactococcus lactis* subsp. *lactis*=*Streptococcus lactis*.

M17 Medium, Modified

Composition per 1001.2mL:

Disodium-β-glycerophosphate.....	9.5g
Pancreatic digest of casein.....	5.0g
Meat peptone.....	5.0g
Papaic digest of soybean.....	5.0g
Yeast extract.....	2.5g
Ascorbic acid.....	0.5g
MgSO ₄ ·7H ₂ O.....	0.25g
Lactose solution.....	50.0mL
CaCl ₂ solution.....	1.2mL

pH 7.15 ± 0.05 at 25°C

Lactose Solution:

Composition per 50.0mL:

Lactose.....	8.0g

Preparation of Lactose Solution: Add lactose to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

CaCl₂ Solution:

Composition per 100.0mL:

CaCl ₂ ·2H ₂ O.....	14.7g

Preparation of CaCl₂ Solution: Add 14.7g of CaCl₂·2H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except lactose solution and CaCl₂ solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 50.0mL of sterile lactose solution and 1.2mL of sterile CaCl₂ solution. Mix thoroughly. Aseptically adjust pH to 7.15 ± 0.05. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Streptococcus thermophilus*.

M9 Medium with Arginine

Composition per 1013.0mL:

L-Proline.....	20.0mg
L-Arginine.....	20.0mg

10X M9 salts.....	100.0mL
Glucose solution.....	10.0mL
CaCl ₂ ·2H ₂ O solution.....	1.0mL
MgSO ₄ solution.....	1.0mL
Thiamine·HCl solution.....	1.0mL
pH 7.4 ± 0.2 at 25°C	

10X M9 Salts:

Composition per liter:

Na ₂ HPO ₄	60.0g
KH ₂ PO ₄	30.0g
NH ₄ Cl.....	10.0g
NaCl.....	5.0g

Preparation of 10X M9 Salts: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4.

Glucose Solution:

Composition per 100.0mL:

Glucose.....	20.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

CaCl₂·2H₂O Solution:

Composition per 100.0mL:

CaCl ₂ ·2H ₂ O.....	1.47g

Preparation of CaCl₂·2H₂O Solution: Add CaCl₂·2H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

MgSO₄ Solution:

Composition per 100.0mL:

MgSO ₄	12.04g

Preparation of MgSO₄ Solution: Add MgSO₄ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Thiamine·HCl Solution:

Composition per 10.0mL:

Thiamine·HCl.....	3.37g

Preparation of Thiamine·HCl Solution: Add thiamine·HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add L-proline, L-arginine, and 10X M9 salts to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile glucose solution, 1.0mL of sterile CaCl₂·2H₂O solution, 1.0mL of sterile MgSO₄ solution, and 1.0mL of sterile thiamine·HCl solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Escherichia coli*.

MACA with Maltose**Composition per liter:**

Yeast extract.....	20.0g
Agar	10.0g
Maltose.....	10.0g
Glucose	10.0g
Proteose peptone No. 3	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	0.1g
pH 6.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus sanfrancisco*.

MacConkey Agar**Composition per liter:**

Pancreatic digest of gelatin.....	17.0g
Agar	13.5g
Lactose.....	10.0g
NaCl.....	5.0g
Bile salts.....	1.5g
Pancreatic digest of casein.....	1.5g
Peptic digest of animal tissue.....	1.5g
Neutral Red.....	0.03g
Crystal Violet	1.0mg
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of coliforms and enteric pathogens based on the ability to ferment lactose. Lactose-fermenting organisms appear as red to pink colonies. Lactose-nonfermenting organisms appear as colorless or transparent colonies.

MacConkey Agar**Composition per liter:**

Peptone.....	20.0g
Agar	12.0g
Lactose	10.0g
Bile salts.....	5.0g

NaCl.....	5.0g
Neutral Red.....	0.075g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of coliforms and enteric pathogens based on the ability to ferment lactose. Lactose-fermenting organisms appear as red to pink colonies. Lactose-nonfermenting organisms appear as colorless or transparent colonies.

**MacConkey Agar, CS
(BAM M91)****Composition per liter:**

Peptone	17.0g
Agar	13.5g
Lactose.....	10.0g
NaCl.....	5.0g
Proteose peptone.....	3.0g
Bile salts.....	1.5g
Neutral Red.....	0.03g
Crystal Violet	1.0mg
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and differentiation of lactose-fermenting and nonfermenting Gram-negative bacteria while also controlling the swarming of *Proteus* species, if present. Lactose-fermenting organisms appear as red to pink colonies. Lactose-nonfermenting organisms appear as colorless or transparent colonies.

**MacConkey Agar, Fluorocult
(Fluorocult MacConkey Agar)****Composition per liter:**

Peptone from casein.....	17.0g
Agar	13.5g
Lactose.....	10.0g

NaCl.....	5.0g
Peptone from meat.....	3.0g
Bile salt mixture.....	1.5g
4-Methylumbelliferyl- β -D-glucuronide.....	0.1g
Neutral Red.....	0.03g
Crystal Violet.....	0.001g

pH 7.1 $\pm$ 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes. The plates are clear and red to red-brown.

Use: For the isolation of *Salmonella*, *Shigella*, and coliform bacteria, in particular *E. coli*, from various materials. The bile salts and crystal violet largely inhibit the growth of Gram-positive microbial flora. Lactose together with the pH indicator neutral red are used to detect lactose-positive colonies and *E. coli* can be seen among these because of fluorescence under UV light.

MacConkey Agar No. 2 (MacConkey II Agar)

Composition per liter:

Peptone.....	20.0g
Agar.....	15.0g
Lactose.....	10.0g
NaCl.....	5.0g
Bile salts No. 2.....	1.5g
Neutral Red.....	0.05g
Crystal Violet.....	1.0mg

pH 7.2 $\pm$ 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of enteric pathogens, especially enterococci, in clinical specimens and in materials of sanitary importance.

MacConkey Agar No. 3

Composition per liter:

Peptone.....	20.0g
Agar.....	15.0g
Lactose.....	10.0g
NaCl.....	5.0g

Bile salts No. 3.....	1.5g
Neutral Red.....	0.03g
Crystal Violet.....	0.001g

pH 7.1 $\pm$ 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of enteric pathogens, especially *Salmonella* and *Shigella*, in clinical specimens and in foods.

MacConkey Agar without Crystal Violet

Composition per liter:

Agar.....	12.0g
Lactose.....	10.0g
Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue.....	10.0g
Bile salts.....	5.0g
NaCl.....	5.0g
Neutral Red.....	0.05g

pH 7.4 $\pm$ 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the detection of members of the Enterobacteriaceae and enterococci as well as some staphylococci. For the isolation and detection of coliforms and enteric pathogens from water and wastewater.

MacConkey Agar without Salt

Composition per liter:

Peptone.....	20.0g
Agar.....	12.0g
Lactose.....	10.0g
Bile salts.....	5.0g
Neutral Red.....	0.075g

pH 7.4 $\pm$ 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat while stirring until boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes. Dry the surface of plates before inoculation.

Use: For the isolation and detection of coliforms and enteric pathogens from urine. Provides a low electrolyte medium on which most *Proteus* species will not swarm and therefore avoids overgrowth of the plate.

MacConkey Broth

Composition per liter:

Pancreatic digest of gelatin.....	20.0g
Lactose.....	10.0g
Oxgall.....	5.0g
Bromcresol Purple.....	0.02g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. If testing 10.0mL samples, prepare medium double strength. Mix thoroughly. Gently heat while stirring until boiling. Distribute into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation and cultivation of coliforms in milk and water.

MacConkey Broth

Composition per liter:

Peptone.....	20.0g
Lactose.....	10.0g
Bile salts.....	5.0g
NaCl.....	5.0g
Neutral Red.....	0.075g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. If testing 10.0mL samples, prepare medium double strength. Mix thoroughly. Gently heat while stirring until boiling. Distribute into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation and cultivation of coliforms in milk and water.

MacConkey Broth, Purple

Composition per liter:

Peptone.....	20.0g
Lactose.....	10.0g

Bile salts.....	5.0g
NaCl.....	5.0g
Bromcresol Purple.....	0.01g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder or tablets from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. If testing 10.0mL samples, prepare medium double strength. Mix thoroughly. Gently heat while stirring until boiling. Distribute into test tubes containing inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation and cultivation of coliforms in milk and water.

Magnesium Oxalate Agar (MOX Agar)

Composition per liter:

Pancreatic digest of casein.....	15.0g
Agar.....	15.0g
Papaic digest of soybean meal.....	5.0g
NaCl.....	5.0g
MgCl ₂ ·6H ₂ O.....	4.1g
Sodium oxalate.....	2.68g
pH 7.4–7.6 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Yersinia enterocolitica* from foods.

Malachite Green Broth

Composition per liter:

Peptone.....	15.0g
Beef extract.....	9.0g
Malachite Green.....	0.01mg
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Pseudomonas aeruginosa*.

Malonate Broth

Composition per liter:

Sodium malonate.....	3.0g
NaCl.....	2.0g

(NH ₄) ₂ SO ₄	2.0g
K ₂ HPO ₄	0.6g
KH ₂ PO ₄	0.4g
Bromthymol Blue	0.025g
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Avoid introduction of carbon and nitrogen from other sources.

Use: For the cultivation and differentiation of coliforms and other enteric organisms, particularly *Enterobacter* and *Escherichia*, based on their ability to utilize malonate as the sole carbon source and ammonium sulfate as the sole nitrogen source. Malonate-utilizing organisms turn the medium blue.

Malonate Broth, Ewing Modified (BAM M92)

Composition per liter:

Sodium malonate	3.0g
NaCl.....	2.0g
(NH ₄) ₂ SO ₄	2.0g
Yeast extract.....	1.0g
Glucose	0.25g
K ₂ HPO ₄	0.6g
KH ₂ PO ₄	0.4g
Bromthymol Blue	0.025g
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of coliforms and other enteric organisms, particularly *Enterobacter* and *Escherichia*, based on their ability to utilize malonate as a carbon source and ammonium sulfate as a nitrogen source. The small amount of yeast extract and glucose encourages the growth of some organisms that may be distressed or fail to respond. Malonate-utilizing organisms turn the medium blue.

Malt Agar

Composition per liter:

Malt extract.....	30.0g
Agar	15.0g
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–118°C. Do not overheat or agar will not harden. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of yeasts and molds.

Malt Agar

Composition per liter:

Agar	20.0g
Malt extract.....	12.5g
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fungi.

Malt Agar (MA) (BAM M185)

Composition per liter:

Agar	20.0g
Malt extract.....	20.0g
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fungi.

Malt and Peptone Medium

Composition per liter:

Agar	15.0g
Malt extract.....	10.0g
Peptone	5.0g
NaCl.....	1.0g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Adjust pH to 6.5. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Flavobacterium* species.

Malt Extract Agar

Composition per liter:

Malt extract	30.0g
Agar	15.0g
Mycological peptone	5.0g
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–115°C. Do not overheat or agar will not harden. If a lower pH (3.5) is desired, cool medium to 55°C and aseptically add 100.0mL of sterile lactic acid. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the detection, isolation, and enumeration of yeasts and molds. The addition of lactic acid suppresses bacterial growth.

Malt Extract Agar

(BAM M93)

Composition per liter:

Malt extract	30.0g
Agar	20.0g
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and identification of heat-resistant filamentous fungi (molds) from foods.

Malt Extract Agar, Half Strength (ATCC Medium 2418)

Composition per liter:

Agar	15.0g
Malt extract	10.0g
Peptone	2.5g
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring until boiling. Distribute into tubes or flasks. Autoclave for 15

min at 15 psi pressure–118°C. Do not overheat or agar will not harden. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of yeasts and molds.

Malt Extract Agar for Yeasts and Molds (MEAYM) (BAM M182)

Composition per liter:

Agar	20.0g
Glucose	20.0g
Malt extract	20.0g
Peptone	1.0g
pH 5.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and identification of heat-resistant filamentous fungi (molds) from foods. Recommended for identification of *Aspergillus* spp. and *Penicillium* spp.

Malt Extract Broth

(BAM M94)

Composition per liter:

Malt extract	6.0g
Glucose	6.0g
Maltose	1.8g
Yeast extract	1.2g
pH 4.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat—this results in darkening of the broth.

Use: For the isolation, cultivation, and enumeration of yeast and filamentous fungi (mold).

Malt Extract Broth

Composition per liter:

Malt extract	17.0g
Mycological peptone	3.0g
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–115°C.

Use: For the cultivation of molds and yeasts, especially for sterility testing.

Malt Extract Broth

Composition per liter:

Malt extract, purified solids 15.0g
pH 4.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–118°C. Do not overheat.

Use: For the cultivation of yeasts and molds.

Malt Extract Yeast Extract 40% Glucose Agar (MY40G)

Composition per liter:

Glucose 400.0g
Agar 12.0g
Malt extract powder 12.0g
Yeast extract 3.0g
pH 5.5 ± 0.2 at 25°C

Preparation of Medium: Add components, except glucose, to 550.0mL of distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Bring volume to 600.0mL with distilled/deionized water. While the solution is still hot, add the glucose all at once while stirring to avoid formation of lumps. Autoclave for 30 min at 0 psi pressure–100°C.

Use: For the isolation and cultivation of osmotolerant microorganisms from foods.

Malt Yeast Extract 50% Glucose Agar (MY50G) (ATCC Medium 2093)

Composition per liter:

Glucose 500.0g
Malt extract 10.0g
Yeast extract 2.5g
Agar 10.0g
pH 5.5 ± 0.2 at 25°C

Preparation of Medium: Add agar, yeast extract, and malt extract to distilled/deionized water and bring volume to 500mL. Mix thoroughly. Gently

heat while stirring until boiling. Slowly add glucose while stirring to avoid lumps. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Do not overheat or agar will not harden. Pour into sterile Petri dishes or leave in tubes. Note: This agar hardens very slowly.

Use: For the cultivation of yeasts and molds.

Manganese Nutrient Agar

Composition per liter:

Agar 15.0g
Peptone 5.0g
Meat extract 3.0g
MnSO₄·H₂O 5.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and obtaining the sporulation of *Bacillus* species.

Mannitol Agar

Composition per liter:

Mannitol 25.0g
Agar 15.0g
Yeast extract 5.0g
Peptone 3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Acetobacter aceti*, *Acetobacter hansenii*, *Acetobacter pasteurianus*, *Frateuria aurantia*, *Gluconobacter asaii*, *Gluconobacter cerinus*, *Gluconobacter oxydans*, and other bacteria that can utilize mannitol as a carbon source.

Mannitol Egg Yolk Polymyxin Agar (MYP Agar)

Composition per liter:

Agar 12.0g
D-Mannitol 10.0g
Peptone 10.0g
NaCl 10.0g
Beef extract 1.0g

Phenol Red	0.025g
Egg yolk emulsion, 50%	50.0mL
Polymyxin B	10.0mL
pH 7.2 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	variable
NaCl (0.85% solution)	40.0mL

Preparation of Egg Yolk Emulsion: Wash fresh eggs with stiff brush and drain. Soak eggs in 70% ethanol for 1 hr. Crack eggs aseptically and separate yolks from whites. Remove egg yolks with a sterile syringe or a wide-mouth pipet. Place 50.0mL of egg yolks into a sterile container. Add 50.0mL sterile 0.85% saline.

Polymyxin B Solution:

Composition per 50.0mL:

Polymyxin B	500,000U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except egg yolk emulsion, 50%, and polymyxin B solution—to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 49°C. Aseptically add 50.0mL of sterile egg yolk emulsion, 50%, and 10.0mL of sterile polymyxin B solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation and enumeration of *Bacillus cereus* from foods.

Mannitol Egg Yolk Polymyxin Agar (MYP Agar) (BAM M95)

Composition per liter:

Agar	15.0g
D-Mannitol	10.0g
Peptone	10.0g
NaCl	10.0g
Beef extract	1.0g
Egg yolk emulsion, 50%	50.0mL
Polymyxin B	10.0mL
Phenol Red, 1% in 95% ethanol	2.5mL
pH 7.2 ± 0.1 at 25°C	

Source: Available as a prepared medium from BD Diagnostic Systems.

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	80.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Polymyxin B Solution:

Composition per 50.0mL:

Polymyxin B	500,000U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except egg yolk emulsion, 50%, and polymyxin B solution—to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile egg yolk emulsion, 50%, and 10.0mL of sterile polymyxin B solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation and enumeration of *Bacillus cereus* from foods.

Mannitol Maltose Agar (BAM M96)

Composition per liter:

NaCl	20.0g
Agar	13.0g
D-Mannitol	10.0g
Maltose	10.0g
Beef extract	5.0g
Papaic digest of soybean meal	5.0g
Polypeptone	5.0g
Dye stock solution, 1000×	1.0mL
pH 7.8 ± 0.2 at 25°C	

Dye Stock Solution, 1000X:

Bromthymol Blue	4.0g
Cresol Red	4.0g
Ethanol, 95%	100.0mL

Preparation of Dye Stock Solution, 1000X:

Add Bromthymol Blue and Cresol Red to 100.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust to pH 7.8. Gently heat and

bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Vibrio* species from foods.

Mannitol Salt Agar

Composition per liter:

NaCl.....	75.0g
Agar	15.0g
D-Mannitol	10.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue.....	5.0g
Beef extract.....	1.0g
Phenol Red.....	0.025g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and enumeration of staphylococci. Mannitol-utilizing organisms turn the medium yellow.

Mannitol Salt Agar (BAM M97)

Composition per liter:

NaCl.....	75.0g
Agar	15.0g
D-Mannitol	10.0g
Polypeptone	10.0g
Beef extract.....	1.0g
Phenol Red.....	0.025g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and enumeration of staphylococci. Mannitol-utilizing organisms turn the medium yellow.

Mannitol Salt Broth

Composition per liter:

NaCl.....	100.0g
D-Mannitol	2.5g

Pancreatic digest casein	17.0g
Papaic digest soy bean meal	3.0g
K ₂ HPO ₄	2.5g
Phenol Red.....	0.025g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation and cultivation of staphylococci from foods. Mannitol-utilizing organisms turn the medium yellow.

Mannitol Yolk Polymyxin Agar (MYP Agar)

Composition per 110.0mL:

Agar	1.5g
NaCl.....	1.0g
Peptone	1.0g
D-Mannitol	1.0g
(NH ₄) ₂ PO ₄	0.1g
Meat extract	0.1g
Phenol Red.....	2.5mg
Egg yolk emulsion, 20%.....	10.0mL
Polymyxin B solution	1.0mL

pH 7.1 ± 0.2 at 25°C

Egg Yolk Emulsion, 20%:

Composition per 100.0mL:

Chicken egg yolks.....	11
Whole chicken egg	1
NaCl (0.9% solution)	80.0mL

Preparation of Egg Yolk Emulsion, 20%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 20.0mL of egg yolk emulsion and add to 80.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Polymyxin B Solution:

Composition per 1.0mL:

Polymyxin B	1.0mg

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 1.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except egg yolk emulsion, 20%, and polymyxin B solution—to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL

of sterile egg yolk emulsion, 20%, and 1.0mL of sterile polymyxin B solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation and maintenance of *Bacillus cereus*.

Maximum Recovery Diluent

Composition per liter:

NaCl.....	8.5g
Peptone.....	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: This diluent is a physiologically isotonic and protective medium for maximal recovery of microorganisms from a variety of sources.

McBride Agar, Modified

Composition per liter:

Agar	15.0g
Glycine anhydride.....	10.0g
Tryptose	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Phenylethanol.....	2.5g
LiCl	0.5g
Cycloheximide solution	10.0mL
pH 7.3 ± 0.2 at 25°C	

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide.....	0.2g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloheximide solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile cycloheximide solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation of *Listeria monocytogenes* from dairy products.

McBride *Listeria* Agar

Composition per liter:

Agar	15.0g
Glycine.....	10.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Phenylethyl alcohol	2.5g
LiCl	0.5g
pH 7.3 ± 0.22 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation of *Listeria monocytogenes* from specimens containing mixed flora.

McClung Carbon-Free Broth

Composition per liter:

NaNO ₃	2.0g
K ₂ HPO ₄	0.8g
MgSO ₄ ·7H ₂ O	0.5g
FeCl ₃	0.01g
MnCl ₂ ·4H ₂ O	8.0mg
ZnSO ₄	2.0mg
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat without boiling until salts dissolve. Cool to 25°C. Adjust pH to 7.2. Filter sterilize.

Use: For use as a basal medium in determining the carbon assimilation capabilities of microorganisms.

McClung-Toabe Agar

Composition per liter:

Proteose peptone.....	40.0g
Agar	25.0g
Na ₂ HPO ₄	5.0g
Glucose	2.0g
NaCl.....	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Egg yolk emulsion, 50%.....	100.0mL
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg.....	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components, except egg yolk emulsion, 50%, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes in 15.0mL volumes.

Use: For the isolation and cultivation of *Clostridium perfringens* in foods.

McClung-Toabe Agar, Modified**Composition per liter:**

Proteose peptone No. 2	40.0g
Agar	20.0g
Na ₂ HPO ₄	5.0g
Glucose	2.0g
NaCl.....	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Egg yolk emulsion, 50%.....	100.0mL
Hemin solution.....	1.0mL

pH 7.6 ± 0.2 at 25°C

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg.....	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Hemin Solution:**Composition per 100.0mL:**

Hemin.....	0.5g
NaOH (1 <i>N</i> solution).....	20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1*N* NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium:

Add components, except egg yolk emulsion, 50%, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of a wide variety of anaerobic bacteria. For the differentiation of anaerobic bacteria based on lecithinase production and lipase production. Bacteria that produce lecithinase appear as colonies surrounded by a zone of insoluble precipitate. Bacteria that produce lipase appear as colonies with a pearly iridescent sheen.

McClung-Toabe Agar, Modified**Composition per liter:**

Proteose peptone No. 2	40.0g
Agar	20.0g
Na ₂ HPO ₄	5.0g
Glucose	2.0g
NaCl.....	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Neomycin.....	0.1g
Egg yolk emulsion, 50%.....	100.0mL
Hemin solution.....	1.0mL

pH 7.6 ± 0.2 at 25°C

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks.....	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Hemin Solution:**Composition per 100.0mL:**

Hemin	0.5g
NaOH (1 <i>N</i> solution).....	20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium: Add components, except egg yolk emulsion, 50%, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Clostridium* species. For the differentiation of *Clostridium* species based on lecithinase production and lipase production. Bacteria that produce lecithinase appear as colonies surrounded by a zone of insoluble precipitate. Bacteria that produce lipase appear as colonies with a pearly iridescent sheen.

McClung-Toabe Agar, Modified

Composition per liter:

Proteose peptone No. 2	20.0g
Agar	20.0g
Yeast extract	5.0g
Pancreatic digest of casein	5.0g
NaCl	5.0g
Sodium thioglycolate	1.0g
Egg yolk emulsion, 50%	80.0mL
pH 7.6 ± 0.2 at 25°C	

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components, except egg yolk emulsion, 50%, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 80.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Clostridium botulinum*.

McClung-Toabe Egg Yolk Agar, CDC Modified (CDC Modified McClung-Toabe Egg Yolk Agar)

Composition per liter:

Pancreatic digest of casein	40.0g
Agar	25.0g
NaHPO ₄	5.0g
Yeast extract	5.0g
D-Glucose	2.0g
NaCl	2.0g
Egg yolk emulsion	100.0mL
MgSO ₄ (5% solution)	0.2mL
pH 7.4 ± 0.2 at 25°C	

Egg Yolk Emulsion:

Composition:

Chicken egg yolks	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs. Separate yolks from whites for 11 eggs. Mix egg yolks with 1 chicken egg.

Preparation of Medium: Add components, except egg yolk emulsion, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 60°C. Aseptically add 100.0mL of sterile egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the isolation, cultivation, and differentiation of anaerobic bacteria from foods. Bacteria that produce lecithinase appear as colonies surrounded by an insoluble opaque precipitate. Bacteria that produce lipase activity appear as colonies with a sheen or “pearly” surface. Bacteria that possess proteolytic activity appear as colonies surrounded by a clear zone.

McClung-Toabe Egg Yolk Agar, CDC Modified (CDC Modified McClung- Toabe Egg Yolk Agar)

Composition per liter:

Pancreatic digest of casein	40.0g
Agar	25.0g
Na ₂ HPO ₄	5.0g
Yeast extract	5.0g
Glucose	2.0g
NaCl	2.0g
KH ₂ PO ₄	1.0g

Egg yolk emulsion, 50%	100.0mL
MgSO ₄ ·7H ₂ O (5% solution)	0.2mL
pH 7.3 ± 0.2 at 25°C	

Egg Yolk Emulsion, 50%:**Composition per 100.0mL:**

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components—except egg yolk emulsion, 50%—to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile egg yolk emulsion, 50%. Mix thoroughly. Pour into sterile Petri dishes in 15.0mL volumes.

Use: For the cultivation of a wide variety of anaerobic bacteria. For the differentiation of anaerobic bacteria based on lecithinase production, lipase production, and proteolytic ability. Bacteria that produce lecithinase appear as colonies surrounded by a zone of insoluble precipitate. Bacteria that produce lipase appear as colonies with a pearly iridescent sheen. Bacteria that produce proteolytic activity appear as colonies surrounded by a clear zone.

m-CP Medium**Composition per liter:**

Tryptose	30.0g
Yeast extract	20.0g
Agar	15.0g
Sucrose	5.0g
L-Cysteine·HCl·H ₂ O	1.0g
MgO ₄ ·7H ₂ O	0.1g
Bromocresol Purple	0.04g
Phenolphthalein biphosphate tetrazolium salt solution	10.0mL
Selective supplement solution	4.0mL
Indoxyl-β-D-glucoside solution	4.0mL
Ferric chloride solution	1.0mL
pH 7.6 ± 0.2 at 25°C	

Selective Supplement Solution:**Composition per 4.0mL:**

D-Cycloserine	0.4g
Polymyxin B sulfate	25.0mg

Preparation of Selective Supplement Solution:

Add components to 4.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Phenolphthalein Biphosphate Tetrazolium**Salt Solution:****Composition per 10.0mL:**

Phenolphthalein biphosphate tetrazolium salt	25.0mg

Preparation of Phenolphthalein Biphosphate

Tetrazolium Salt Solution: Add phenolphthalein biphosphate tetrazolium salt to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Indoxyl-β-D-glucoside Solution:**Composition per 10.0mL:**

Indoxyl-β-D-glucoside	0.45g

Preparation of Indoxyl-β-D-glucoside Solution:

Add indoxyl-β-D-glucoside to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Ferric Chloride Solution:**Composition per 4.0mL:**

FeCl ₃ ·6H ₂ O	30.0mg

Preparation of Ferric Chloride Solution:

Add FeCl₃·6H₂O to 4.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components except selective supplement solution, phenolphthalein biphosphate tetrazolium salt solution, indoxyl-β-D-glucoside solution, and ferric chloride solution, to distilled/deionized water and bring volume to 981.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 4.0mL of selective supplement solution. Mix thoroughly. Aseptically add 10.0mL phenolphthalein biphosphate tetrazolium salt solution, 4.0mL indoxyl-β-D-glucoside solution, and 1.0mL ferric chloride solution. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into tubes.

Use: A selective, chromogenic medium for the rapid identification and enumeration of *Clostridium perfringens* in water samples, including water used in food and beverage production.

MD Medium**Composition per liter:**

Agar	20.0g
L-Malic acid	20.0g
Pancreatic digest of casein	10.0g
D-Glucose	5.0g
Casamino acids	3.0g
Pancreatic digest of soybean meal	1.5g
Tween 80	1.0g

Yeast extract.....	1.0g
Bromcresol Green solution.....	20.0mL
pH 7.0 ± 0.2 at 25°C	

Bromcresol Green Solution:**Composition per 30.0mL:**

Bromcresol Green.....	0.1g
NaOH (0.01 <i>N</i> solution).....	30.0mL

Preparation of Bromcresol Green Solution:

Add Bromcresol Green to 30.0mL of NaOH solution. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Adjust pH to 7.0 with 10*N* KOH. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Salmonella* species from foods.

Medium for Aciduric, Thermophilic *Bacillus* Strains

Composition per liter:

Solution A.....	500.0mL
Solution B.....	500.0mL
pH 4.3 ± 0.2 at 25°C	

Solution A:**Composition per 500.0mL:**

KH ₂ PO ₄	3.0g
Glucose.....	1.0g
Starch.....	1.0g
Tryptone.....	1.0g
Yeast extract.....	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
CaCl ₂ ·2H ₂ O.....	0.25g
(NH ₄) ₂ SO ₄	0.2g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Adjust pH to 4.3. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution B:**Composition per 500.0mL:**

Agar.....	20.0g

Preparation of Solution B: Add agar to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Preparation of Medium: Aseptically combine 500.0mL of solution A and 500.0mL of solution B. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of aciduric, thermophilic *Bacillus* strains.

Medium for Halophilic Bacilli**Composition per liter:**

NaCl.....	100.0g
Casamino acids.....	10.0g
Yeast extract.....	10.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of halophilic *Bacillus* species.

Medium for Lactobacilli (ATCC Medium 980)

Composition per liter:

Agar.....	20.0g
Peptone.....	12.5g
Glucose.....	11.0g
Sodium acetate.....	10.0g
Yeast extract.....	5.5g
KH ₂ PO ₄	0.25g
K ₂ HPO ₄	0.25g
MgSO ₄	0.1g
MnSO ₄ ·4H ₂ O.....	0.05g
FeSO ₄ ·7H ₂ O.....	0.05g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.8. Autoclave for 10 min at 15 psi pressure–120°C. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Pedio-coccus acidilactici* and *Bacillus* species.

Medium N**Composition per liter:**

Agar.....	20.0g
Glucose.....	20.0g
Yeast nitrogen base without amino acids.....	6.7g
Casamino acids, vitamin free.....	2.0g
Isoleucine.....	0.1g
Valine.....	0.1g
Deoxythymidine-5'-monophosphate solution.....	10.0mL

Deoxythymidine-5'-Monophosphate Solution:**Composition per 10mL:**

Deoxythymidine-5'-monophosphate.....	15.0mg

Preparation of Deoxythymidine-5'-Monophosphate Solution: Add deoxythymidine-5'-monophosphate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except deoxythymidine-5'-monophosphate solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 60°C. Aseptically add 10.0mL of sterile deoxythymidine-5'-monophosphate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

Medium S

Composition per liter:

Glucose	10.0g
K ₂ HPO ₄	4.0g
Peptone	4.0g
Yeast extract	4.0g
KH ₂ PO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.5g
pH 7.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the general cultivation of a wide variety of bacteria.

Membrane *Clostridium perfringens* Medium (m-CP Medium)

Composition per 1040mL:

Tryptose	30.0g
Yeast extract	20.0g
Agar	15.0g
Sucrose	5.0g
L-Cysteine·HCl·H ₂ O	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Bromocresol Purple	0.04g
Phenolphthalein solution	20.0mL
Indoxyl-β-D-glucoside solution	8.0mL
Selective supplement solution	8.0mL
Ferric chloride solution	4.0mL
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Selective Supplement Solution:

Composition per 8.0mL:

D-Cycloserine	0.8g
Polymyxin B sulfate	50.0mg

Preparation of Selective Supplement Solution: Add components to distilled/deionized water and bring volume to 8.0mL. Mix thoroughly. Filter sterilize.

Indoxyl-β-D-glucoside Solution:

Composition per 10.0mL:

Indoxyl-β-D-glucoside	75.0mg

Preparation of Indoxyl-β-D-glucoside Solution: Add indoxyl-β-D-glucoside to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Ferric Chloride Solution:

Composition per 10.0mL:

FeCl ₃ ·6H ₂ O	0.45g

Preparation of Ferric Chloride Solution: Add ferric chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Phenolphthalein Solution:

Composition per 20.0mL:

Phenolphthalein biphosphate tetrasodium salt	0.15g

Preparation of Phenolphthalein Solution: Add phenolphthalein biphosphate tetrasodium salt to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except selective supplement solution, phenolphthalein solution, ferric chloride solution, and indoxyl-β-D-glucoside to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 8.0mL selective supplement solution, 20.0mL phenolphthalein solution, 4.0mL ferric chloride solution, and 8.0mL indoxyl-β-D-glucoside. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the rapid isolation and presumptive identification of *Clostridium perfringens* from food and water samples. A selective and chromogenic medium for the presumptive identification of *Clostridium perfringens*, especially from water samples. Recommended in European Council Directive 98/83/EC for testing the quality of water intended for human consumption. *C. perfringens* colonies have a characteristic opaque yellow appearance. Most other *Clostridium* spp. will appear as either purple colonies, due to the lack of sucrose fermentation, or blue/green colo-

nies where the organism is still cleaving indoxyl- β -D-glucoside and also fermenting sucrose.

Membrane Lactose Glucuronide Agar (MLGA)

Composition per liter:

Peptone.....	40.0g
Lactose	30.0g
Agar	10.0g
Yeast extract.....	6.0g
Sodium lauryl sulfate.....	1.0g
Sodium pyruvate.....	0.5g
5-Bromo-4-chloro-3-indoxyl- β -D-glucuronic acid.....	0.2g
Phenol Red.....	0.2g
pH 7.4 $\pm$ 0.2 at 25°C	

Source: This medium is available from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the direct enumeration of *E. coli* and coliforms in foods by the membrane filtration method. The chromogenic substrate 5-Bromo-4-chloro-3-indoxyl- β -D-glucuronic acid (BCIG), is cleaved by the enzyme β -glucuronidase and produces a blue chromophore that builds up within the bacterial cells. In addition, the incorporation of phenol red detects lactose fermentation and results in yellow colonies when acid is produced. Since coliform colonies are lactose positive, they will appear yellow on this medium and as *E. coli* colonies are both lactose and β -glucuronidase positive, they will appear green.

Membrane Lauryl Sulfate Broth

Composition per liter:

Peptone.....	39.0g
Lactose	30.0g
Yeast extract.....	6.0g
Sodium lauryl sulfate.....	1.0g
Phenol Red.....	0.2g
pH 7.4 $\pm$ 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enumeration of coliform organisms and *Escherichia coli*.

Methylene Blue Milk Medium (MBM Medium)

Composition per liter:

Skim milk, dehydrated.....	100.0g
Methylene Blue.....	10.0g
pH 6.4 $\pm$ 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 20 min at 10 psi pressure–115°C.

Use: For the cultivation and differentiation of group D streptococci (enterococci) from other *Streptococcus* species.

MIB with Maltose

Composition per liter:

Yeast extract.....	20.0g
Maltose	10.0g
Glucose	10.0g
Proteose peptone No. 3	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	0.1g
pH 6.7 $\pm$ 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus sanfrancisco*.

Micro Assay Culture Agar

Composition per liter:

Yeast extract.....	20.0g
Agar	10.0g
Glucose	10.0g
Proteose peptone No. 3	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	0.1g
pH 6.7 $\pm$ 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Just prior to solidification of the agar, disperse precipitate by gently twirling tube.

Use: For carrying stock cultures of lactobacilli and other test microorganisms used in microbiological assays. For the general cultivation of lactobacilli.

Micro Inoculum Broth**Composition per liter:**

Yeast extract.....	20.0g
Glucose	10.0g
Proteose peptone No. 3	5.0g
KH ₂ PO ₄	2.0g
Sorbitan monooleate complex	0.1g
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of lactobacilli used in microbiological assays. It is of particular value in the preparation of the inoculum for these tests.

Microbial Content Test Agar**Composition per liter:**

Agar	15.0g
Pancreatic digest of casein.....	15.0g
NaCl.....	5.0g
Tween 80.....	5.0g
Enzymatic hydrolysate of soybean meal	5.0g
Lecithin	0.7g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Boil for 1–2 min. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For determining the efficiency of sanitization of containers, equipment, and environmental surfaces.

Micrococcus Medium**Composition per liter:**

Agar	15.0g
Peptone.....	5.0g
Yeast extract.....	3.0g
Beef extract.....	1.5g
Glucose	1.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Staphylococcus aureus* and *Micrococcus* species.

Micrococcus Medium, FDA**Composition per liter:**

Agar	15.0g
Proteose peptone.....	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Staphylococcus aureus* and *Micrococcus* species.

Middlebrook 13A Medium**Composition per 112.5mL:**

Casein hydrolysate.....	0.1g
Tween 80.....	0.02g
Sodium polyanetholesulfonate.....	0.025g
Catalase.....	36,000U
¹⁴ C-substrate	125μCi (185kBq)
Middlebrook 7H9 broth	100.0mL
Middlebrook 13A enrichment.....	12.5mL
pH 6.6 ± 0.2 at 25°C	

Middlebrook 7H9 Broth:**Composition per liter:**

Na ₂ HPO ₄	2.5g
KH ₂ PO ₄	1.0g
Monosodium glutamate	0.5g
(NH ₄) ₂ SO ₄	0.5g
Sodium citrate.....	0.1g
MgSO ₄ ·7H ₂ O	0.05g
Ferric ammonium citrate.....	0.04g
CuSO ₄ ·5H ₂ O.....	1.0mg
Pyridoxine.....	1.0mg
ZnSO ₄ ·7H ₂ O	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Glycerol	2.0mL

Preparation of Middlebrook 7H9 Broth: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Middlebrook 13A Enrichment:**Composition per 20.0mL:**

Bovine serum albumin.....	3.0g

Preparation of Middlebrook 13A Enrichment: Add bovine serum albumin to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 100.0mL of Middlebrook 7H9 broth, add remaining components, except Middlebrook 13A enrichment. Mix thoroughly. Filter sterilize. Aseptically distribute into bottles in 4.0mL volumes. Prior to inoculation, aseptically add 0.5mL of Middlebrook 13A enrichment to each bottle. Mix thoroughly.

Use: For the cultivation of *Mycobacterium* species from the blood of patients suspected of having mycobacteremia.

Middlebrook ADC Enrichment (Middlebrook Albumin Dextrose Catalase Enrichment)

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
Catalase	0.003g

Source: This medium is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Use: For use as a supplement to other Middlebrook media for the isolation, cultivation, and maintenance of *Mycobacterium* species. Also used as a supplement to other Middlebrook media for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H9 Broth with Middlebrook ADC Enrichment

Composition per liter:

Na ₂ HPO ₄	2.5g
KH ₂ PO ₄	1.0g
Monosodium glutamate	0.5g
(NH ₄) ₂ SO ₄	0.5g
Sodium citrate	0.1g
MgSO ₄ ·7H ₂ O	0.05g
Ferric ammonium citrate	0.04g
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
ZnSO ₄ ·7H ₂ O	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Middlebrook ADC enrichment	100.0mL
Glycerol	2.0mL

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook ADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
Catalase	3.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook ADC Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook ADC enrichment. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook ADC enrichment. Mix thoroughly. Distribute into sterile tubes or flasks.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H9 Broth with Middlebrook OADC Enrichment

Composition per liter:

Na ₂ HPO ₄	2.5g
KH ₂ PO ₄	1.0g
Monosodium glutamate	0.5g
(NH ₄) ₂ SO ₄	0.5g
Sodium citrate	0.1g
MgSO ₄ ·7H ₂ O	0.05g
Ferric ammonium citrate	0.04g
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
ZnSO ₄ ·7H ₂ O	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Middlebrook OADC enrichment	100.0mL
Glycerol	2.0mL

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook OADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g

Oleic acid	0.05g
Catalase	4.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook OADC Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook OADC enrichment. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment. Mix thoroughly. Distribute into sterile tubes or flasks.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H9 Broth with Middlebrook OADC Enrichment and Triton WR 1339

Composition per liter:

Na ₂ HPO ₄	2.5g
KH ₂ PO ₄	1.0g
Monosodium glutamate	0.5g
(NH ₄) ₂ SO ₄	0.5g
Sodium citrate	0.1g
MgSO ₄ ·7H ₂ O	0.05g
Ferric ammonium citrate	0.04g
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
ZnSO ₄ ·7H ₂ O	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Middlebrook OADC enrichment with Triton WR 1339	100.0mL
Glycerol	2.0mL
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook OADC Enrichment with Triton WR 1339:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Triton WR 1339	0.25g
Oleic acid	0.05g
Catalase	4.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook OADC Enrichment with Triton WR 1339: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook OADC enrichment with Triton WR-1339. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment with Triton WR-1339. Mix thoroughly. Distribute into sterile tubes or flasks.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H10 Agar with Middlebrook ADC Enrichment

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
(NH ₄) ₂ SO ₄	0.5g
L-Glutamic acid	0.5g
Sodium citrate	0.4g
Ferric ammonium citrate	0.04g
MgSO ₄ ·7H ₂ O	0.025g
ZnSO ₄ ·7H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Malachite Green	0.25mg
Middlebrook ADC enrichment	100.0mL
Glycerol	5.0mL
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook ADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
Catalase	0.003g
Distilled water	100.0mL

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook ADC Enrichment: Add components to distilled/deionized water

and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook ADC enrichment. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook ADC enrichment. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H10 Agar with Middlebrook OADC Enrichment (Middlebrook and Cohn 7H10 Agar)

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
(NH ₄) ₂ SO ₄	0.5g
L-Glutamic acid	0.5g
Sodium citrate	0.4g
Ferric ammonium citrate	0.04g
MgSO ₄ ·7H ₂ O	0.025g
ZnSO ₄ ·7H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Malachite Green	0.25mg
Middlebrook OADC enrichment	100.0mL
Glycerol	5.0mL

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook OADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Oleic acid	0.05g
Catalase	4.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook OADC Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook OADC enrichment. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H10 Agar with Middlebrook OADC Enrichment and Hemin (Hemin Medium for *Mycobacterium*)

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
(NH ₄) ₂ SO ₄	0.5g
L-Glutamic acid	0.5g
Sodium citrate	0.4g
Ferric ammonium citrate	0.04g
MgSO ₄ ·7H ₂ O	0.025g
ZnSO ₄ ·7H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Malachite Green	0.25mg
Middlebrook OADC enrichment	100.0mL
Glycerol	5.0mL
Hemin solution	3.9mL

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook OADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Oleic acid	0.05g
Catalase	4.0mg

Preparation of Middlebrook OADC Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Hemin Solution:

Composition per 100.0mL:

Hemin	1.0g
NaOH (1N solution)	20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium: Add glycerol to 891.1mL of distilled/deionized water and add remaining components, except Middlebrook OADC enrichment. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. For the cultivation and maintenance of *Mycobacterium haemophilum*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook 7H10 Agar with Middlebrook OADC Enrichment and Triton WR 1339

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
(NH ₄) ₂ SO ₄	0.5g
L-Glutamic acid	0.5g
Sodium citrate	0.4g
Ferric ammonium citrate	0.04g
MgSO ₄ ·7H ₂ O	0.025g
ZnSO ₄ ·7H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O	0.5mg
Malachite Green	0.25mg
Middlebrook OADC enrichment with Triton WR 1339	100.0mL
Glycerol	5.0mL
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Middlebrook OADC Enrichment with Triton WR 1339:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Triton WR 1339	0.25g
Oleic acid	0.05g
Catalase	4.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook OADC Enrichment with Triton WR 1339: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water and add remaining components, except Middlebrook OADC enrichment with Triton WR-1339. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment with Triton WR-1339. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and maintenance of *Mycobacterium* species, including *Mycobacterium tuberculosis*. Also used for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook OADC Enrichment (Middlebrook Oleic Albumin Dextrose Catalase Enrichment)

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Oleic acid	0.05g
Catalase	4.0mg

Source: This medium is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Use: For use as a supplement to other Middlebrook media for the isolation, cultivation, and maintenance of *Mycobacterium* species. Also used as a supplement to other Middlebrook media for determining the antimicrobial susceptibility of mycobacteria.

Middlebrook OADC Enrichment with Triton WR 1339 (Middlebrook Oleic Albumin Dextrose Catalase Enrichment with Triton WR 1339)

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl	0.85g
Triton WR 1339	0.25g

Oleic acid	0.05g
Catalase	4.0mg

Source: This medium is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Use: For use as a supplement to other Middlebrook media for the isolation, cultivation, and maintenance of *Mycobacterium* species. Also used as a supplement to other Middlebrook media for determining the antimicrobial susceptibility of mycobacteria.

MIL Medium (Motility Indole Lysine Medium)

Composition per liter:

Peptone	10.0g
Pancreatic digest of casein	10.0g
L-Lysine·HCl	10.0g
Yeast extract	3.0g
Agar	2.0g
Dextrose	1.0g
Ferric ammonium citrate	0.5g
Bromocresol Purple	0.02g

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder and prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of members of the *Enterobacteriaceae* on the basis of motility, lysine decarboxylase activity, lysine deaminase activity, and indole production.

Milk Agar

Composition per liter:

Agar	15.0g
Peptone	5.0g
Yeast extract	3.0g
Milk solids or	1.0g
fresh milk	10.0mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of microorganisms from dairy samples.

Minerals Modified Glutamate Agar

Composition per liter:

Agar	15.0g
Lactose	10.0g
Sodium glutamate	6.35g
NH ₄ Cl	2.5g
K ₂ HPO ₄	0.9g
Sodium formate	0.25g
MgSO ₄ ·7H ₂ O	0.1g
L-Aspartic acid	0.024g
L-Arginine	0.02g
L-Cystine	0.02g
Bromocresol Purple	0.01g
CaCl ₂ ·2H ₂ O	0.01g
Ferric ammonium citrate	0.01g
Nicotinic acid	1.0mg
Pantothenic acid	1.0mg
Thiamine	1.0mg

pH 6.7 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 11 psi pressure–116°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of coliform bacteria from foods.

Minerals Modified Medium

Composition per liter:

Lactose	20.0g
Sodium glutamate	12.7g
NH ₄ Cl	5.0g
K ₂ HPO ₄	1.8g
Sodium formate	0.5g
MgSO ₄ ·7H ₂ O	0.2g
L-Aspartic acid	0.048g
L-Cystine	0.04g
L-Arginine	0.04g
Ferric ammonium citrate	0.02g
CaCl ₂ ·2H ₂ O	0.02g
Bromocresol Purple	0.02g
Thiamine	2.0mg
Nicotinic acid	2.0mg
Pantothenic acid	2.0mg

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add NH_4Cl to distilled/deionized water and bring volume to 800.0mL. Add remaining components and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.7. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–116°C. Check pH after autoclaving. This medium is double strength.

Use: For the enumeration of coliform bacteria.

Minimal Agar I

Composition per liter:

Agar	20.0g
Glucose	20.0g
$(\text{NH}_4)_2\text{SO}_4$	5.0g
KH_2PO_4	1.0g
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	0.5g
NaCl	0.1g
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	0.1g
Inositol	2.0mg
KI	1.0mg
H_3BO_3	0.5mg
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.4mg
$\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$	0.4mg
Thiamine-HCl	0.4mg
Pyroxidine-HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl_3	0.2mg
$\text{Na}_2\text{MoO}_4 \cdot 4\text{H}_2\text{O}$	0.2mg
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.04mg
Folic acid	2.0 μg
Biotin	2.0 μg
Growth supplement solution	10.0mL
pH 5.5 $\pm$ 0.2 at 25°C	

Growth Supplement Solution:

Composition per 10.0mL:

L-Tryptophan	20.0mg
Uracil	20.0mg
L-Histidine-HCl	20.0mg

Preparation of Growth Supplement Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Minimal Agar II

Composition per liter:

Agar	20.0g
Glucose	20.0g
$(\text{NH}_4)_2\text{SO}_4$	5.0g
KH_2PO_4	1.0g
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	0.5g
NaCl	0.1g
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	0.1g
Inositol	2.0mg
KI	1.0mg
H_3BO_3	0.5mg
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	0.4mg
$\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$	0.4mg
Thiamine-HCl	0.4mg
Pyroxidine-HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl_3	0.2mg
$\text{Na}_2\text{MoO}_4 \cdot 4\text{H}_2\text{O}$	0.2mg
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	0.04mg
Folic acid	2.0 μg
Biotin	2.0 μg
Growth supplement solution	10.0mL
pH 5.5 $\pm$ 0.2 at 25°C	

Growth Supplement Solution:

Composition per 10.0mL:

Adenine sulfate	20.0mg
L-Arginine-HCl	20.0mg
L-Histidine-HCl	20.0mg

Preparation of Growth Supplement Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Minimal Agar III

Composition per liter:

Agar	20.0g
Glucose	20.0g

(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
Inositol.....	2.0mg
KI.....	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyroxidine·HCl.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.2mg
CuSO ₄ ·5H ₂ O.....	0.04mg
Folic acid.....	2.0μg
Biotin.....	2.0μg
Growth supplement solution.....	10.0mL
pH 5.5 ± 0.2 at 25°C	

Growth Supplement Solution:

Composition per 10.0mL:

L-Leucine.....	30.0mg
L-Tryptophan.....	20.0gm
Uracil.....	20.0mg

Preparation of Growth Supplement Solution:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Minimal Broth I

Composition per liter:

Glucose.....	20.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
Inositol.....	2.0mg
KI.....	1.0mg
H ₃ BO ₃	0.5mg

ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyroxidine·HCl.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.2mg
CuSO ₄ ·5H ₂ O.....	0.04mg
Folic acid.....	2.0μg
Biotin.....	2.0μg
Growth supplement solution.....	10.0mL

pH 5.5 ± 0.2 at 25°C

Growth Supplement Solution:

Composition per 10.0mL:

L-Tryptophan.....	20.0mg
Uracil.....	20.0mg
L-Histidine·HCl.....	20.0mg

Preparation of Growth Supplement Solution:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Minimal Broth II

Composition per liter:

Glucose.....	20.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
Inositol.....	2.0mg
KI.....	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyroxidine·HCl.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg

Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg
Growth supplement solution.....	10.0mL
pH 5.5 ± 0.2 at 25°C	

Growth Supplement Solution:**Composition per 10.0mL:**

Adenine sulfate	20.0mg
L-Arginine·HCl	20.0mg
L-Histidine·HCl.....	20.0mg

Preparation of Growth Supplement Solution:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Minimal Broth III**Composition per liter:**

Agar	20.0g
Glucose	20.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine·HCl	0.4mg
Pyroxidine·HCl	0.4mg
Niacin.....	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg
Growth supplement solution.....	10.0mL
pH 5.5 ± 0.2 at 25°C	

Growth Supplement Solution:**Composition per 10.0mL:**

L-Leucine	30.0mg
L-Tryptophan	20.0mg
Uracil	20.0mg

Preparation of Growth Supplement Solution:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium:

Add components, except growth supplement solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of sterile growth supplement solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Saccharomyces cerevisiae*.

MLCB Agar**(Mannitol Lysine Crystal Violet-Brilliant Green Agar)****Composition per liter:**

Agar	15.0g
Peptone	10.0g
Yeast extract.....	5.0g
L-Lysine·HCl.....	5.0g
NaCl.....	4.0g
Na ₂ S ₂ O ₃	4.0g
Mannitol.....	3.0g
Beef extract.....	2.0g
Ferric ammonium citrate.....	1.0g
Crystal Violet	0.01g
Brilliant Green	12.5mg
pH 6.8 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the selective isolation and cultivation of *Salmonella* species from foods.

**Modified *Campylobacter* Blood-Free
Selective Agar Base
(Modified *Campylobacter* Charcoal
Differential Agar)
(Modified CCDA)
(BAM M30a)**

Composition per 1012mL:

Agar	12.0g

Beef extract.....	10.0g
Peptone.....	10.0g
NaCl.....	5.0g
Charcoal.....	4.0g
Casein hydrolysate.....	3.0g
Yeast extract.....	2.0g
Sodium deoxycholate.....	1.0g
FeSO ₄	0.25g
Sodium pyruvate.....	0.25g
Cefoperazone solution.....	4.0mL
Amphotericin B solution.....	4.0mL
Rifampicin solution.....	4.0mL

pH 7.4 ± 0.2 at 25°C

Cefoperazone Solution:

Composition per 10.0mL:

Cefoperazone.....	0.037g

Preparation of Cefoperazone Solution: Add cefoperazone to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Rifampicin Solution:

Composition per 100.0mL:

Rifampicin.....	0.25g
Ethanol, absolute.....	50.0mL

Preparation of Rifampicin Solution: Add rifampicin to 50.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Amphotericin B Solution:

Composition per 10.0mL:

Amphotericin B.....	0.005g

Preparation of Amphotericin B Solution: Add Amphotericin B to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize. Can be stored for 1 year at -20°C.

Preparation of Medium: Add components, except cefoperazone solution, amphotericin B solution, and rifampicin solution to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure-121°C. Cool to 45°-50°C. Aseptically add 10.0mL of sterile cefoperazone solution, 10.0mL of sterile amphotericin B solution, and 10.0mL of sterile rifampicin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Campylobacter* species. For the recovery of injured *Campylobacter* spp. from foods.

Modified Oxford *Listeria* Selective Agar (Oxford Agar, Modified) (*Listeria* Selective Agar, Modified Oxford) (MOX Agar) (BAM M103a)

Composition per 1002mL:

Special peptone.....	23.0g
LiCl.....	15.0g
Agar.....	12.0g
NaCl.....	5.0g
Cornstarch.....	1.0g
Esculin.....	1.0g
Ferric ammonium citrate.....	0.5g
Buffered colistin methane sulfonate solution.....	1.0mL
Buffered moxalactam solution.....	2.0mL

pH 7.2 ± 0.2 at 25°C

Buffered Colistin Methane Sulfonate Solution:

Composition per 100.0mL:

Colistin methane sulfonate.....	1.0g
Potassium phosphate buffer, 0.1M. pH 6.0. 100.0mL	

Preparation of Buffered Colistin Methane Sulfonate Solution: Add colistin methane sulfonate to 100.0mL 0.1M potassium phosphate buffer. Mix thoroughly. Adjust pH to 6.0. Filter sterilize. Store at -20°C.

Buffered Moxalactam Solution:

Composition per 100.0mL:

Sodium or ammonium moxalactam.....	1.0g

Preparation of Buffered Moxalactam Solution: Add sodium or ammonium moxalactam to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Store at -20°C.

Preparation of Medium: Gradually add components, except buffered moxalactam solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Autoclave for 10 min at 15 psi pressure-121°C. Cool quickly to 46°C. Aseptically add 2.0mL of sterile moxalactam solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes* from specimens containing a mixed bacterial flora.

Motility Nitrate Medium (BAM M101)

Composition per liter:

Tryptose.....	10.0g
Agar.....	3.0g

Beef Heart, Infusion from 500g.....	2.0g
Tryptose	2.0g
NaCl.....	1.0g
KNO ₃	1.0g
Glucose	0.5g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with agitation to dissolve agar. Distribute into screw-cap tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the detection of motility in Gram-negative nonfermentative bacteria.

Modified Semisolid Rappaport Vassiliadis Medium (MSRV Medium)

Composition per liter:

MgCl ₂ , anhydrous	10.93g
NaCl.....	7.34g
Casein hydrolysate.....	4.59g
Tryptose	4.59g
Agar	2.7g
KH ₂ PO ₄	1.47g
Malachite Green oxalate	0.037g
Novobiocin.....	10.0mL

pH 5.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Novobiocin Solution:

Composition per 10.0mL:

Novobiocin.....	0.02g

Preparation of Novobiocin Solution: Add novobiocin to 10.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat to boiling. Do not autoclave. Cool to 45°–50°C. Aseptically add 10.0mL of sterile novobiocin solution. Mix thoroughly. Pour into sterile Petri dishes. Air-dry plates for at least 1 hr.

Use: For the isolation and cultivation of motile *Salmonella* species from food samples.

Møller Decarboxylase Broth

Composition per liter:

Amino acid.....	10.0g
Peptic digest of animal tissue.....	5.0g

Beef extract.....	5.0g
Glucose	0.5g
Bromcresol Purple	0.01g
Cresol Red	5.0mg
Pyridoxal.....	5.0mg

pH 6.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Use L-lysine, L-arginine, or L-ornithine. Mix thoroughly. Gently heat until dissolved. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. A slight precipitate may form in the ornithine broth.

Use: For the differentiation of Gram-negative enteric bacteria based on the production of arginine dihydrolase, lysine decarboxylase, or ornithine decarboxylase.

Møller KCN Broth Base

Composition per liter:

Na ₂ HPO ₄	5.64g
NaCl.....	5.0g
Pancreatic digest of casein.....	1.5g
Peptic digest of animal tissue	1.5g
KH ₂ PO ₄	0.225g
KCN solution	0.15mL

pH 7.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

KCN Solution:

Composition per 100.0mL:

KCN.....	0.5g

Preparation of KCN Solution: Add KCN to 100.0mL of cold distilled/deionized water. Mix thoroughly and cap. Do not mouth pipette.

Caution: Cyanide is toxic.

Preparation of Medium: Add components, except KCN solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature. Prior to use, add 0.15mL of KCN solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the differentiation of Gram-negative enteric bacteria on the basis of their ability to grow in the presence of cyanide.

Motility Indole Ornithine Medium (MIO Medium)

Composition per liter:

Pancreatic digest of gelatin.....	10.0g

Pancreatic digest of casein.....	9.5g
L-Ornithine·HCl	5.0g
Yeast extract.....	3.0g
Agar	2.0g
Glucose	1.5g
Bromcresol Purple	0.02g
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of Gram-negative enteric bacteria based on their motility, indole production, and ornithine decarboxylase activity.

Motility Indole Ornithine Medium (MIO Medium) (BAM M99)

Composition per liter:

Tryptone.....	10.0g
Peptone.....	10.0g
L-Ornithine·HCl	5.0g
Yeast extract.....	3.0g
Agar	2.0g
Glucose	1.0g
Bromcresol Purple	0.02g
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of Gram-negative enteric bacteria based on their motility, indole production, and ornithine decarboxylase activity.

Motility Medium (BAM M100)

Composition per liter:

Pancreatic digest of casein.....	10.0g
Glucose	5.0g
Agar	3.0g
Na ₂ HPO ₄	2.5g
Yeast extract.....	2.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 2.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to stand at 25°C for 2 days prior to inoculation.

Use: For the cultivation and observation of motility of *Bacillus cereus*.

Motility Medium S

Composition per liter:

Beef heart, solids from infusion.....	500.0g
Gelatin.....	30.0g
Enzymatic hydrolyzate of protein.....	10.0g
NaCl.....	5.0g
K ₂ HPO ₄	2.0g
KNO ₃	2.0g
Agar	1.0g
2,3,5-Triphenyltetrazolium chloride solution	10.0mL
pH 7.2 ± 0.2 at 25°C	

2,3,5-Triphenyltetrazolium Chloride Solution:

Composition per 10.0mL:

2,3,5-Triphenyltetrazolium chloride	0.1g

Preparation of 2,3,5-Triphenyltetrazolium Chloride Solution: Add 2,3,5-triphenyltetrazolium chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except 2,3,5-triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 60°C. Aseptically add 10.0mL of the sterile 2,3,5-triphenyltetrazolium chloride solution. Mix thoroughly. Aseptically distribute into sterile tubes. Keep at 4°–8°C until used.

Use: For the determination of bacterial motility.

Motility Nitrate Agar

Composition per liter:

Beef heart, solids from infusion.....	100.0g
Tryptose	12.0g
Agar	3.0g
NaCl.....	1.0g
KNO ₃	1.0g
Glucose	0.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and observation of motility and nitrate reduction in a variety of Gram-negative bacteria.

Motility Nitrate Medium (FDA M101)

Composition per liter:

Beef heart, solids from infusion.....	100.0g
Tryptose	12.0g
Agar	3.0g
NaCl.....	1.0g
KNO ₃ , nitrite free.....	1.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of Gram-negative nonfermentative bacteria from cosmetics based on their motility and their ability to reduce nitrate to nitrite.

Motility Nitrate Medium, Buffered (BAM M102)

Composition per liter:

Peptone.....	5.0g
Galactose.....	5.0g
Agar	3.0g
Beef extract.....	3.0g
Na ₂ HPO ₄	2.5g
KNO ₃	1.0g
Glycerin	5.0mL
pH 7.3 ± 0.1 at 25°C	

Preparation of Medium: Add components, except agar, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Add agar. Gently heat until boiling. Distribute into tubes in 11.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. If not used within 4 hr, heat tubes to 100°C for 10 min.

Use: For the cultivation and observation of the motility of *Clostridium perfringens*.

Motility Sulfide Medium

Composition per liter:

Gelatin.....	80.0g
Proteose peptone.....	10.0g
NaCl.....	5.0g

Agar	4.0g
Beef extract.....	3.0g
Sodium citrate.....	2.0g
L-Cystine	0.2g
Ferrous ammonium citrate.....	0.2g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes in 4–5.0mL volumes. Autoclave for 15 min at 10 psi pressure–116°C.

Use: For the determination of bacterial motility and the ability of bacteria to produce H₂S from L-cystine. For the differentiation of Gram-negative bacteria of the *Enterobacteriaceae*.

Motility Test and Maintenance Medium Composition per liter:

Peptone	10.0g
NaCl.....	5.0g
Agar	4.0g
Beef extract.....	3.0g
2,3,5-Triphenyltetrazolium chloride.....	0.05g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 8.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation, maintenance, and observation of the motility of *Listeria monocytogenes*.

Motility Test and Maintenance Medium

Composition per liter:

Agar	9.0g
Tryptose	8.0g
NaCl.....	5.0g
Pancreatic digest of gelatin.....	2.5g
Beef extract.....	1.5g
pH 7.2 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 7.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pass the cooled tubes into an anaerobic chamber containing 85% N₂ + 10% H₂ + 5% CO₂.

Use: For the cultivation, maintenance, and observation of the motility in a variety of anaerobic bacteria.

Motility Test and Maintenance Medium

Composition per liter:

Peptone.....	10.0g
NaCl.....	5.0g
Agar.....	4.0g
Beef extract.....	3.0g

pH 7.4 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 8.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation, maintenance, and observation of motility in members of the *Enterobacteriaceae*.

Motility Test and Maintenance Medium, Gilardi

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
Agar.....	3.0g
Yeast extract.....	3.0g

pH 7.2 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 3.5mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation, maintenance, and observation of motility in nonfermenting Gram-negative bacteria.

Motility Test and Maintenance Medium, Tatum

Composition per liter:

Tryptose.....	8.0g
NaCl.....	5.0g
Agar.....	4.0g
Pancreatic digest of gelatin.....	2.5g
Beef extract.....	1.5g

pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 8.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation, maintenance, and observation of motility in nonfermenting Gram-negative bacteria.

Motility Test Medium

Composition per liter:

Pancreatic digest of gelatin.....	10.0g
NaCl.....	5.0g
Agar.....	4.0g
Beef extract.....	3.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

2,3,5-Triphenyltetrazolium Chloride Solution:

Composition per 10.0mL:

2,3,5-Triphenyltetrazolium chloride.....	0.1g

Preparation of 2,3,5-Triphenyltetrazolium Chloride Solution: Add 2,3,5-triphenyltetrazolium chloride to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 995.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.0mL of sterile 2,3,5-triphenyltetrazolium chloride solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For detection of the motility of Gram-negative enteric bacteria.

Motility Test Medium

Composition per liter:

Tryptose.....	10.0g
NaCl.....	5.0g
Agar.....	5.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes in 4–5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes quickly in an upright position.

Use: For the determination of bacterial motility.

Motility Test Medium, Semisolid with NaCl (BAM M103)

Composition per liter:

NaCl.....	25.0g
Peptone.....	10.0g

Agar	4.0g
Beef extract	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into screw-capped tubes in 8.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Leave in tubes.

Use: For the cultivation and observation of motility in a variety of bacteria, especially *Salmonella* spp. and *Listeria* spp. For *Listeria* spp. keep tubes tightly screw capped and sealed with parafilm. For *Salmonella* spp. use Petri plates prepared on the same day as use.

Motility Test Medium, Semisolid

Composition per liter:

Peptone	10.0g
NaCl	5.0g
Agar	4.0g
Beef extract	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into screw-capped tubes in 8.0mL or 20.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes or leave in tubes.

Use: For the cultivation and observation of motility in a variety of bacteria, especially *Salmonella* species.

Motility Test Medium, Semisolid (BAM M103)

Composition per liter:

Peptone	10.0g
NaCl	5.0g
Agar	4.0g
Beef extract	3.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into screw-capped tubes in 8.0mL or 20.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes or leave in tubes.

Use: For the cultivation and observation of motility in halophilic *Vibrio* spp.

MP 5 Medium (Mineral Pectin 5 Medium)

Composition per liter:

Agar solution	500.0mL
Basal medium	250.0mL
Mineral solution	250.0mL
pH 5.0–6.0 at 25°C	

Agar Solution:

Composition per 500.0mL:

Agar	15.0g

Preparation of Agar Solution: Add agar to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Basal Medium:

Composition per 250.0mL:

Na ₂ HPO ₄	6.0g
Pectin, citrus or apple	5.0g
KH ₂ PO ₄	4.0g
NH ₄ SO ₄	2.0g
Yeast extract	1.0g

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 250.0mL. Mix thoroughly. Gently heat and bring to boiling.

Mineral Solution:

Composition per 250.0mL:

FeSO ₄ (0.1% solution)	1.0mL
MgSO ₄ ·7H ₂ O (20% solution)	1.0mL
CaCl ₂ ·2H ₂ O (0.1% solution)	1.0mL
H ₃ BO ₃ (0.001% solution)	1.0mL
MnSO ₄ ·H ₂ O (0.001% solution)	1.0mL
ZnSO ₄ ·7H ₂ O (0.007% solution)	1.0mL
CuSO ₄ ·5H ₂ O (0.005% solution)	1.0mL
MoO ₃ (0.001% solution)	1.0mL

Preparation of Mineral Solution: Add components to distilled/deionized water and bring volume to 250.0mL. Mix thoroughly.

Preparation of Medium: Combine 250.0mL of basal medium and 250.0mL of mineral solution. Mix thoroughly. Adjust pH to 5.0–6.0 with 1N HCl. Autoclave the basal medium–mineral solution and agar solution separately for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically combine the two sterile solutions. Mix thoroughly. Pour immediately into sterile Petri dishes to prevent hydrolysis of the agar.

Use: For the cultivation of microorganisms that produce polygalactanase.

MP 7 Medium (Mineral Pectin 7 Medium)

Composition per liter:

Basal medium	500.0mL
Mineral solution	500.0mL
pH 7.2 ± 0.2 at 25°C	

Basal Medium:

Composition per 500.0mL:

Agar	15.0g
Na ₂ HPO ₄	6.0g
Pectin, citrus or apple	5.0g
KH ₂ PO ₄	4.0g
NH ₄ SO ₄	2.0g
Yeast extract	1.0g

Preparation of Basal Medium: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling.

Mineral Solution:

Composition per 500.0mL:

FeSO ₄ (0.1% solution)	1.0mL
MgSO ₄ ·7H ₂ O (20% solution)	1.0mL
CaCl ₂ ·2H ₂ O (0.1% solution)	1.0mL
H ₃ BO ₃ (0.001% solution)	1.0mL
MnSO ₄ ·H ₂ O (0.001% solution)	1.0mL
ZnSO ₄ ·7H ₂ O (0.007% solution)	1.0mL
CuSO ₄ ·5H ₂ O (0.005% solution)	1.0mL
MoO ₃ (0.001% solution)	1.0mL

Preparation of Mineral Solution: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Preparation of Medium: Combine 500.0mL of basal medium and 500.0mL of mineral solution. Mix thoroughly. Adjust pH to 7.2. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Pour into sterile Petri dishes.

Use: For the cultivation of microorganisms that produce pectate lyase.

MPH Agar (Milk Protein Hydrolysate Agar)

Composition per liter:

Agar	15.0g
Casein hydrolysate	9.0g
Glucose	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring

to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically distribute into sterile tubes. Cool to 43°–45°C before using.

Use: For use in the enumeration of bacteria in dairy products.

MRS Agar (DeMan, Rogosa, Sharpe Agar)

Composition per liter:

Glucose	20.0g
Peptone	10.0g
Agar	10.0g
Beef extract	8.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	4.0g
K ₂ HPO ₄	2.0g
Triammonium citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Sorbitan monooleate	1.0mL
pH 6.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of lactic acid bacteria.

MRS Agar (DeMan, Rogosa, Sharpe Agar)

Composition per liter:

Glucose	18.5g
Agar	13.5g
Pancreatic digest of gelatin	10.0g
Beef extract	8.0g
Yeast extract	4.0g
Sodium acetate	3.0g
K ₂ HPO ₄	2.0g
Ammonium citrate	2.0g
Polysorbate 80	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
pH 6.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave

for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Lactobacillus* species from foods and dairy products.

MRS Agar with Cysteine (LMG Medium 118)

Composition per liter:

Glucose	20.0g
Peptone	10.0g
Agar	12.0g
Beef extract	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg
L-Cysteine solution	10.0mL
Tween 80	1.0mL

pH 6.5 ± 0.2 at 25°C

L-Cysteine Solution:

Composition per 10.0mL:

L-Cysteine	0.5g

Preparation of L-Cysteine Solution: Add 0.1g of L-cysteine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteine solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 6.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL of sterile L-cysteine solution. Mix thoroughly. Aseptically pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Leuconostoc mesenteroides* and other lactic acid bacteria.

MRS Agar with Ethanol (LMG Medium 130)

Composition per liter:

Glucose	20.0g
Agar	12.0g
Peptone	10.0g
Beef extract	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg

Ethanol	100.0mL
Tween 80	1.0mL
pH 5.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except ethanol, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 100.0mL of sterile ethanol. Mix thoroughly. Aseptically distribute into sterile tubes or pour into sterile Petri dishes.

Use: For the cultivation of *Lactobacillus acetotolerans*.

MRS Agar with Tomato Juice pH 5.2 (LMG Medium 248)

Composition per liter:

Glucose	20.0g
Peptone	10.0g
Agar	12.0g
Beef extract	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg
Tomato juice	100.0mL
Tween 80	1.0mL

pH 5.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Lactobacillus kunkeei*.

MRS Agar, Half Strength (LMG Medium 281)

Composition per liter:

Glucose	10.0g
Agar	6.0g
Peptone	5.0g
Beef extract	2.5g
Sodium acetate·3H ₂ O	2.5g
Yeast extract	2.5g
KH ₂ PO ₄	1.0g
Diammonium hydrogen citrate	1.0g
MgSO ₄ ·7H ₂ O	0.1g
MnSO ₄ ·4H ₂ O	19.0mg
Tween 80	0.5mL

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus acidipiscis* and *Weissella thailandensis*.

MRS Broth (LMG Medium 166)

Glucose	20.0g
Peptone.....	10.0g
Beef extract.....	5.0g
Sodium acetate·3H ₂ O.....	5.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg
Sorbitan monooleate	1.0mL
pH 6.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Leuconostoc pseudomesenteroides*, *Leuconostoc carnosum*, and *Lactobacillus crispatus*.

MRS Broth (DeMan, Rogosa, Sharpe Broth)

Composition per liter:

Glucose	20.0g
Peptone.....	10.0g
Beef extract.....	8.0g
Sodium acetate·3H ₂ O.....	5.0g
Yeast extract.....	4.0g
K ₂ HPO ₄	2.0g
Triammonium citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Sorbitan monooleate	1.0mL
pH 6.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of lactic acid bacteria.

MRS Broth (DeMan, Rogosa, Sharpe Broth)

Composition per liter:

Glucose	18.5g
Pancreatic digest of gelatin.....	10.0g
Beef extract.....	8.0g
Yeast extract.....	4.0g
Sodium acetate.....	3.0g
K ₂ HPO ₄	2.0g
Ammonium citrate.....	2.0g
Polysorbate 80	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g

pH 6.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Lactobacillus* species from foods and dairy products.

MRS Broth with CaCO₃ (LMG Medium 166)

CaCO ₃	30.0g
Glucose	20.0g
Peptone	10.0g
Beef extract.....	5.0g
Sodium acetate·3H ₂ O.....	5.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg
Sorbitan monooleate	1.0mL

pH 6.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Sporolactobacillus inulinus*.

MRS Broth with Cysteine

Composition per liter:

Glucose	18.5g
Pancreatic digest of gelatin.....	10.0g
Beef extract.....	8.0g
Yeast extract.....	4.0g
Sodium acetate.....	3.0g
K ₂ HPO ₄	2.0g

Ammonium citrate	2.0g
Polysorbate 80	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
L-Cysteine solution	0.5mL
pH 6.2 ± 0.2 at 25°C	

L-Cysteine Solution:**Composition** per 10.0mL:

L-Cysteine	0.1g

Preparation of L-Cysteine Solution: Add 0.1g of L-cysteine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteine solution, to distilled/deionized water and bring volume to 999.5mL. Mix thoroughly. Adjust pH to 6.2. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 0.5mL of sterile L-cysteine solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and cultivation of *Leuconostoc mesenteroides*.

MRS Broth with Ethanol (LMG Medium 240)

Composition per liter:

Glucose	20.0g
Peptone	10.0g
Beef extract	5.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	38.0mg
Ethanol	100.0mL
Tween 80	1.0mL
pH 5.0± 0.2 at 25°C	

Preparation of Medium: Add components, except ethanol, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 100.0mL of sterile ethanol. Mix thoroughly. Aseptically distribute into sterile tubes or sterile flasks.

Use: For the cultivation of *Lactobacillus acetotolerans*.

MRS Broth with Ethanol**Composition** per liter:

Glucose	20.0g
Peptone	10.0g
Beef extract	8.0g
Sodium acetate·3H ₂ O	5.0g

Yeast extract	4.0g
K ₂ HPO ₄	2.0g
Triammonium citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Sorbitan monooleate	1.0mL
Ethanol (95% solution), filter sterilized	100.0mL
pH 5.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except ethanol, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 100.0mL of sterile ethanol. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Lactobacillus acetotolerans*.

MRS Chalk**Composition** per liter:

CaCO ₃	30.0g
Glucose	20.0g
Beef extract	10.0g
Peptone	10.0g
Yeast extract	5.0g
K ₂ HPO ₄	2.0g
Triammonium citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Tween 80	1.0mL
pH 6.2 ± 0.4 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Sporolactobacillus inulinus* and other *Sporolactobacillus* species.

MRS Medium (DSMZ Medium 11)

Composition per liter:

Glucose	20.0g
Casein peptone, tryptic digest	10.0g
Meat extract	10.0g
Yeast extract	5.0g
Na-acetate	5.0g
K ₂ HPO ₄	2.0g
(NH ₄) ₂ citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·2H ₂ O	0.05g
Tween 80	1.0mL
pH 6.2–6.5	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2–6.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus* spp., *Leuconostoc mesenteroides*, and *Pediococcus pentosaceus*.

MRS with Fermented Wort

Composition per liter:

Solution A	800.0mL
Solution B	200.0mL
pH 6.2 ± 0.2 at 25°C	

Solution A:

Composition per liter:

Glucose	20.0g
Peptone.....	10.0g
Agar	10.0g
Beef extract	8.0g
Sodium acetate·3H ₂ O	5.0g
Yeast extract	4.0g
K ₂ HPO ₄	2.0g
Triammonium citrate	2.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
Sorbitan monooleate	1.0mL

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution B:

Composition per liter:

Malt extract	15.0g
Maltose.....	12.75g
Dextrin	2.75g
Glycerol	2.35g
K ₂ HPO ₄	1.0g
NH ₄ Cl	1.0g
Pancreatic digest of gelatin	0.78g

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to room temperature. Inoculate with *Saccharomyces cerevisiae* to allow wort to ferment. Incubate for 24–48 hr. Allow to cool to room temperature. Filter through Whatman filter paper to remove solids. Filter sterilize.

Preparation of Medium: Aseptically combine 800.0mL of sterile solution A and 200.0mL of sterile

solution B. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of yeasts and fungi.

MRVP Broth

(Methyl Red-Voges-Proskauer Broth) (BAM M104 Medium 1)

Composition per liter:

Buffered peptone-water powder	7.0g
Glucose	5.0g
KH ₂ PO ₄	5.0g
pH 7.0 ± 0.2 at 25°C	

Source: Buffered peptone-water powder is available from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of bacteria based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

MRVP Broth

(Methyl Red-Voges-Proskauer Broth) (BAM M104 Medium 2)

Composition per liter:

Glucose	5.0g
KH ₂ PO ₄	5.0g
Pancreatic digest of casein.....	3.5g
Peptic digest of animal tissue	3.5g
pH 6.9 ± 0.2 at 25°C	

Source: Available as a premixed powder from BD Diagnostic Systems and as a prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of bacteria based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

MRVP Medium

(BAM M104 Medium 3)

(Methyl Red-Voges-Proskauer Medium)

Composition per liter:

Glucose	5.0g
Peptone	5.0g
Phosphate buffer	5.0g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of bacteria based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

MRVP Broth with NaCl
(Methyl Red-Voges-Proskauer
Broth with NaCl)
(BAM M104 Medium 1)

Composition per liter:

NaCl	30.0g
Buffered peptone-water powder	7.0g
Glucose	5.0g
KH ₂ PO ₄	5.0g
pH 7.0 ± 0.2 at 25°C	

Source: Buffered peptone-water powder is available from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of halophilic *Vibrio* spp. based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

MRVP Broth with NaCl
(Methyl Red-Voges-Proskauer
Broth with NaCl)
(BAM M104 Medium 2)

Composition per liter:

NaCl	30.0g
Glucose	5.0g
KH ₂ PO ₄	5.0g
Pancreatic digest of casein	3.5g
Peptic digest of animal tissue	3.5g
pH 6.9 ± 0.2 at 25°C	

Source: Available as a premixed powder from BD Diagnostic Systems and as a prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of halophilic *Vibrio* spp. based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

MRVP Broth with NaCl
(Methyl Red- Voges-Proskauer
Broth with NaCl)
(Methyl Red Voges-Proskauer Medium)
Composition per liter:

NaCl	30.0g
Glucose	5.0g
Peptone	5.0g
Phosphate buffer	5.0g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of halophilic *Vibrio* spp. based on acid production (Methyl Red test) and acetoin production (Voges-Proskauer reaction).

Mucate Broth
(BAM M105)

Composition per liter:

Mucic acid	10.0g
Peptone	10.0g
Bromthymol Blue	0.024g
pH 7.4 ± 0.1 at 25°C.	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Add 5N NaOH while stirring until mucic acid dissolves. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of enterovirulent *Escherichia coli* and *Shigella* species.

Mucate Control Broth
(BAM M106)

Composition per liter:

Peptone	10.0g
Bromthymol Blue	0.024g
pH 7.4 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 5.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of enterovirulent *Escherichia coli* and *Shigella* species.

Mueller-Hinton Agar (BAM M107)

Composition per liter:

Beef infusion from.....	300.0g
Acid hydrolysate of casein.....	17.5g
Agar	17.0g
Starch	1.5g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostics and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 11 psi pressure–116°C. Pour into sterile Petri dishes or leave in tubes.

Use: For antimicrobial susceptibility testing of a variety of bacterial species.

Mueller-Hinton Agar with Horse Blood (LMG Medium 49)

Composition per liter:

Beef infusion from.....	300.0g
Acid hydrolysate of casein.....	17.5g
Agar	17.0g
Starch	1.5g
Horse blood, sterile defibrinated.....	50.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium without horse blood is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components, except horse blood, to 950.0mL distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL sterile horse blood. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Campylobacter* spp. and other bacteria.

Mueller-Hinton Agar with NaCl (BAM M107)

Composition per liter:

Beef infusion from.....	300.0g
NaCl.....	30.0g
Acid hydrolysate of casein.....	17.5g

Agar	17.0g
Starch	1.5g
pH 7.3 ± 0.2 at 25°C	

Source: This medium without NaCl is available as a premixed powder from BD Diagnostics and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 11 psi pressure–116°C. Pour into sterile Petri dishes or leave in tubes.

Use: For antimicrobial susceptibility testing of a variety of halophilic *Vibrio* spp.

MY10-12 Medium (DSMZ Medium 982)

Composition per liter:

Glucose.....	120.0g
NaCl.....	100.0g
Malt extract.....	20.0g
Agar	20.0g
Yeast extract.....	5.0g
pH 6.7 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Sterilize by steaming for 30 min. Overheating will cause softening. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of osmophilic yeasts and fungi.

MY20 Agar

Composition per liter:

Glucose.....	200.0g
Agar	20.0g
Peptone	5.0g
Yeast extract.....	3.0g
Malt extract.....	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of osmophilic heterotrophic bacteria and fungi.

MY40 Agar

Composition per liter:

Sucrose.....	400.0g
Agar	20.0g

Malt extract.....	20.0g
Yeast extract.....	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of osmophilic heterotrophic bacteria and fungi.

MY60 Agar

Composition per liter:

Sucrose.....	600.0g
Agar.....	20.0g
Malt extract.....	20.0g
Yeast extract.....	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of osmophilic heterotrophic bacteria and fungi.

Mycobactosel™ Agar

Composition per liter:

Agar.....	13.5g
Bovine albumin fraction V.....	5.0g
Glucose.....	2.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
Pancreatic digest of casein.....	1.0g
NaCl.....	0.85g
(NH ₄) ₂ SO ₄	0.5g
Monosodium glutamate.....	0.5g
Sodium citrate.....	0.4g
MgSO ₄ ·7H ₂ O.....	0.05g
Ferric ammonium citrate.....	0.04g
Pyridoxine.....	1.0mg
ZnSO ₄ ·7H ₂ O.....	1.0mg
CuSO ₄ ·5H ₂ O.....	1.0mg
Biotin.....	0.5mg
CaCl ₂ ·H ₂ O.....	0.5mg
Malachite Green.....	0.25mg
Antibiotic solution.....	10.0mL
Catalase solution.....	10.0mL
Glycerol.....	5.0mL
Oleic acid.....	0.06mL

pH 6.6 ± 0.2 at 25°C

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide.....	0.36g
Nalidixic acid.....	0.02g
Lincomycin.....	2.0mg

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Catalase Solution:

Composition per 10.0mL:

Catalase.....	3.0mg

Preparation of Catalase Solution: Add catalase to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution and catalase solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution and sterile catalase solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of mycobacteria from specimens containing mixed flora.

Mycological Broth

Composition per liter:

Glucose.....	40.0g
Enzymatic hydrolysate of soybean meal.....	10.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fungi.

Mycological Broth with Low pH

Composition per liter:

Glucose.....	40.0g
Enzymatic hydrolysate of soybean meal.....	10.0g

pH 4.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Adjust pH to 4.8. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of saprophytic species of yeasts and molds. It is also suitable for cultivation of aciduric bacteria.

Mycophil Agar

Composition per liter:

Agar	16.0g
Papaic digest of soybean meal	10.0g
Glucose	10.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation, maintenance, and enumeration of fungi. For the demonstration of pigment production in fungal species. Also used for the cultivation and maintenance of *Bacillus* species.

Mycophil Agar with Low pH

Composition per liter:

Agar	18.0g
Papaic digest of soybean meal	10.0g
Glucose	10.0g

pH 4.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Adjust pH to 4.7 by adding approximately 10.0mL of sterile 10% lactic acid. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and enumeration of yeasts and molds.

Mycophil Broth

Composition per liter:

Glucose	40.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–118°C. Do not overheat.

Use: For the isolation and cultivation of a wide variety of fungi.

Mycosel™ Agar

(Cycloheximide Chloramphenicol Agar)

Composition per liter:

Agar	15.5g
Papaic digest of soybean meal	10.0g
Glucose	10.0g
Cycloheximide	0.4g
Chloramphenicol	0.05g

pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 14 psi pressure–118°C. Avoid overheating. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of pathogenic fungi from specimens with other fungi and bacteria.

Neopeptone Glucose Agar

Composition per liter:

Agar	20.0g
Glucose	10.0g
Neopeptone	5.0g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Adjust pH to 6.5. Pour into sterile Petri dishes or leave in tubes.

Use: For the maintenance of stock cultures of a variety of microorganisms.

Neopeptone Glucose Rose Bengal Aureomycin® Agar

Composition per liter:

Agar	20.0g
Neopeptone	5.0g
Glucose	1.0g

Tetracycline solution.....	5.0mL
Rose Bengal solution	3.5mL
pH 6.5 ± 0.2 at 25°C	

Tetracycline Solution:

Composition per 150.0mL:

Tetracycline.....	1.0g

Preparation of Tetracycline Solution: Add tetracycline to distilled/deionized water and bring volume to 150.0mL. Mix thoroughly. Filter sterilize.

Rose Bengal Solution:

Composition per 100.0mL:

Rose Bengal	1.0g

Preparation of Rose Bengal Solution: Add Rose Bengal to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except tetracycline solution, to distilled/deionized water and bring volume to 995.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.0mL of sterile tetracycline solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of a wide variety of fungal species.

Niacin Assay Medium

Composition per liter:

Glucose	40.0g
Sodium acetate.....	20.0g
Vitamin assay casamino acids.....	12.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
L-Cystine	0.4g
MgSO ₄ ·7H ₂ O	0.4g
DL-Tryptophan.....	0.2g
Adenine sulfate	0.02g
FeSO ₄ ·5H ₂ O.....	0.02g
Guanine·HCl	0.02g
MnSO ₄ ·H ₂ O	0.02g
NaCl.....	0.02g
Uracil	0.02g
Pyridoxine·HCl	0.4mg
Riboflavin	0.4mg
Calcium pantothenate	0.2mg
Thiamine·HCl	0.2mg
p-Aminobenzoic acid.....	0.1mg
Biotin	0.8μg

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 5.0mL volumes. Add standard solution or test solutions to each tube. Adjust the volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the microbiological assay of nicotinic acid or nicotinamide (niacin) using *Lactobacillus plantarum* as the test organism.

Nickels and Leesment Agar, Modified

Composition per liter:

Part 1	750.0mL
Part 2	100.0mL
Part 3	100.0mL
Part 4	50.0mL

pH 6.65 ± 0.2 at 25°C

Part 1:

Composition per 750.0mL:

Pancreatic digest of casein.....	20.0g
Lactose	10.0g
Yeast extract.....	5.0g
NaCl.....	4.0g
Gelatin.....	2.5g
Sodium citrate.....	2.0g

Preparation of Part 1: Add components to distilled/deionized water and bring volume to 750.0mL. Mix thoroughly. Adjust pH to 6.65.

Part 2:

Composition per 500.0mL:

Nonfat dry milk.....	50.0g

Preparation of Part 2: Add nonfat dry milk to distilled/deionized water and bring volume to 500.0mL. Inoculate with *Lactobacillus bulgaricus*. Incubate at 20°C for 24 hr. Centrifuge at 5000 rpm for 10 min to separate the curd. Collect the supernatant solution. Autoclave for 15 min at 15 psi pressure–121°C. Store at 4°C.

Part 3:

Composition per 100.0mL:

Calcium citrate.....	13.3g
Carboxymethylcellulose	0.8g

Preparation of Part 3: Combine the calcium citrate and carboxymethylcellulose in a mortar and grind until a fine powder. Add powder to 100.0mL of hot distilled/deionized water. Mix thoroughly. Filter through cheesecloth.

Part 4:

Composition per 50.0mL:

Calcium lactate	8.0g

Preparation of Part 4: Add calcium lactate to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Gently heat until dissolved.

Preparation of Medium: Prepare each of the four parts separately. Autoclave each part for 15 min at 15 psi pressure–121°C. Aseptically combine part 1, part 2, part 3, and part 4. Mix thoroughly. Pour into sterile Petri dishes. Swirl flask while dispensing medium.

Use: For the isolation and cultivation of acid-producing microorganisms from foods.

Nitrate Agar

Composition per liter:

Agar	12.0g
Peptone.....	5.0g
Beef extract.....	3.0g
KNO ₃	1.0g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Broth (International Streptomyces Project Medium 8) (ISP Medium 8) (ATCC Medium 872) (BAM M108)

Composition per liter:

Peptone.....	5.0g
Beef extract.....	3.0g
KNO ₃	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate. Test for nitrates with sulfanilic acid and

α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Broth

Composition per liter:

Pancreatic digest of gelatin.....	20.0g
KNO ₃	2.0g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Broth, *Campylobacter*

Composition per liter:

Beef heart, solids from infusion.....	500.0g
Tryptose	10.0g
NaCl.....	5.0g
KNO ₃	2.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0. Distribute 4.0mL volumes into test tubes that contain inverted Durham tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of *Campylobacter* species based on their ability to reduce nitrate.

Nitrate Broth, Enriched

Composition per liter:

Pancreatic digest of casein.....	13.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Heart muscle, solids from infusion.....	2.0g
KNO ₃	2.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate to nitrite or form N₂ gas. Test

for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Broth, Enriched (BAM M109)

Composition per liter:

Pancreatic digest of casein	13.0g
NaCl	5.0g
Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
KNO ₃	2.0g

pH 7.3 $\pm$ 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Broth

Composition per liter:

Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
KNO ₃	1.0g

pH 6.9 $\pm$ 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of members of the *Pseudomonadaceae* based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Broth

Composition per liter:

Pancreatic digest of casein	13.0g
NaCl	5.0g
Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
KNO ₃ or NaNO ₃	2.0g

pH 7.4 $\pm$ 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of a variety of Gram-negative bacteria based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Broth

Composition per liter:

Pancreatic digest of casein	13.0g
NaCl	5.0g
Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
KNO ₃ or NaNO ₃	2.0g

pH 7.4 $\pm$ 0.2 at 25°C

Preparation of Medium: Dispense in 3.0mL amounts into screw-capped tubes, add inverted vials, and autoclave at 121°C for 15 min.

Use: For the differentiation of a variety of nonfermenting Gram-negative bacteria based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Broth, Clark

Composition per liter:

Peptone	20.0g
KNO ₃ or NaNO ₃	2.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of a variety of Gram-negative bacteria based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α -naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Broth for *Pseudomonas* and Related Genera

Composition per liter:

Peptone	5.0g
NaCl	5.0g
Yeast extract	2.0g

Beef extract.....	1.0g
NaNO ₃	0.1g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of members of the *Pseudomonadaceae* based on their ability to reduce nitrate to nitrite or form N₂ gas. Test for nitrates with sulfanilic acid and α-naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nitrate Reduction Medium and Reagents (BAM M110)

Composition per liter:

Peptone.....	5.0g
Beef extract.....	3.0g
KNO ₃	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Reagent A:

Composition per 100mL:

α-Naphthylamine.....	0.5g
Acetic acid, 5 <i>N</i>	100.0mL

Caution: α-Naphthylamine is a carcinogen.

Preparation of Reagent A: Prepare 5*N* acetic acid by adding distilled/deionized water to 28.7mL glacial acetic acid to give a final volume of 100.0mL. Add 0.5g α-naphthylamine to 100.0mL 5*N* acetic acid. Mix thoroughly.

Reagent B:

Composition per 100mL:

Sulfanilic acid.....	0.8g
Acetic acid, 5 <i>N</i>	100.0mL

Preparation of Reagent B: Prepare 5*N* acetic acid by adding distilled/deionized water to 28.7mL glacial acetic acid to give a final volume of 100.0mL. Add 0.8g sulfanilic acid to 100.0mL 5*N* acetic acid. Mix thoroughly. Gently heat while mixing.

Reagent C:

Composition per 200mL:

α-Naphthol.....	1.0g
Acetic acid.....	200.0mL

Preparation of Reagent C: Add 1.0g α-naphthol to 200.0mL acetic acid. Mix thoroughly.

Zinc Powder:

Cadmium Reagent:

Preparation of Cadmium Reagent: Place zinc rods in 20% solution of CdSO₄. Allow to stand for several hours. Draw off precipitated cadmium and add to 1*N* HCl.

Use: For the differentiation of aerobic and facultative Gram-negative microorganisms based on their ability to reduce nitrate. Test for nitrates with sulfanilic acid and α-naphthylamine reagents. Bacteria that reduce nitrate to nitrite turn the reagents red or pink.

Nonfat Dry Milk, Reconstituted (BAM M111)

Composition per liter:

Nonfat dry milk.....	100.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Swirl until dissolved. Distribute into test tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Salmonella* spp. and for the growth of monkey kidney cells for viral culture.

Novobiocin Agar

Composition per liter:

Agar.....	15.0g
Peptone.....	5.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g
Novobiocin solution.....	10.0mL

Novobiocin Solution:

Composition per 10.0mL:

Novobiocin.....	10.0mg

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 10.0mL of

sterile novobiocin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Staphylococcus aureus*.

**Nutrient Agar
(LMG Medium 160)**

Composition per liter:

Agar	3.0g
Lab-Lemco beef extract.....	1.0g
Peptone.....	1.0g
NaCl.....	0.5g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of heterotrophic bacteria.

Nutrient Agar

Composition per liter:

Agar	15.0g
Peptone.....	5.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of microorganisms.

**Nutrient Agar
(ATCC Medium 3)
(BAM M112)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave

for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a wide variety of bacteria.

**Nutrient Agar
(BAM M113)**

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostics.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into screw-capped tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Pour into sterile Petri dishes or leave in tubes. For slants place tubes in an inclined position while allowing agar to solidify.

Use: For the cultivation of *Bacillus cereus*.

**Nutrient Agar, 1.5%
(ATCC Medium 105)**

Composition per liter:

Agar	15.0g
NaCl.....	8.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of nonfastidious bacteria.

Nutrient Agar No.2

Composition per liter:

Agar	20.0g
Meat extract	10.0g
Peptone	10.0g
NaCl.....	5.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of bacteria.

Nutrient Agar, pH 6.0

Composition per liter:

Agar	15.0g
Peptone.....	5.0g
Beef extract.....	3.0g
pH 6.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of microorganisms that prefer a slightly acid nutrient agar.

Nutrient Agar with 0.5% NaCl

Composition per liter:

Agar	15.0g
NaCl.....	5.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: Nutrient agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Agrobacterium tumefaciens*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella choleraesuis*, *Shigella dysenteriae*, *Shigella flexneri*, *Vibrio* species, and *Yersinia* species.

Nutrient Agar with 0.5% NaCl, pH 9.5-10.0 (LMG Medium 253)

Composition per liter:

Agar	15.0g
NaCl.....	10.0g

Peptone	5.0g
Yeast extract.....	2.0g
Lab-Lemco beef extract.....	1.0g
pH 9.5-10.0	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Adjust pH to 9.5-10.0 with sterile Na₂CO₃ solution. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* spp.

Nutrient Agar with 0.5% NaCl and Sodium Citrate

Composition per liter:

Agar	15.0g
NaCl.....	5.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
Sodium citrate.....	2.94g
pH 6.8 ± 0.2 at 25°C	

Source: Nutrient agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

Nutrient Agar with 3% NaCl

Composition per liter:

NaCl.....	30.0g
Agar	15.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: Nutrient agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* species, *Alteromonas nigrifaciens*, *Halococcus* spe-

cies, *Planococcus citreus*, *Pseudomonas beijerinckii*, *Staphylococcus* species, *Streptococcus pyogenes*, and *Vibrio* species.

Nutrient Agar with Phytone

Composition per liter:

Agar	15.0g
Phytone	10.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a wide variety of bacteria.

Nutrient Agar with Potato Starch

Composition per liter:

Agar	15.0g
Potato starch	10.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: Nutrient agar is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus polymyxa* and *Bacillus subtilis*.

Nutrient Agar with 2% Sucrose (ATCC Medium 1297)

Composition per liter:

Sucrose	20.0g
Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of osmophilic bacteria

Nutrient Broth (BAM M114)

Composition per liter:

Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of nonfastidious microorganisms.

Nutrient Broth No. 2

Composition per liter:

Beef extract	10.0g
Peptone	10.0g
NaCl	5.0g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of fastidious and nonfastidious microorganisms.

Nutrient Broth Salts Medium

Composition per liter:

Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
NaCl	3.0g
MgSO ₄ ·7H ₂ O	0.2g
CaCl ₂ ·2H ₂ O	0.15g
MnSO ₄ ·H ₂ O	0.05g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Bacillus subtilis*.

Nutrient Broth, Standard II

Composition per liter:

Special peptone	8.6g
NaCl	6.4g
pH 7.5 ± 0.1 at 37°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of fastidious and nonfastidious microorganisms.

Nutrient Gelatin

Composition per liter:

Gelatin	120.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring to 50°C. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to liquefy gelatin.

Nutrient Gelatin

Composition per liter:

Beef heart, solids from infusion	500.0g
Gelatin	120.0g
Tryptose	10.0g
NaCl	5.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of Gram-negative, nonfermentative bacteria from foods.

Nutrient Gelatin, CDC (BAM M115)

Composition per liter:

Gelatin	120.0g
Pancreatic digest of casein	13.0g
NaCl	5.0g

Yeast extract	5.0g
Heart muscle, solids from infusion	2.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 55°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of Gram-negative, nonfermentative bacteria from foods.

NZC Broth

Composition per liter:

N-Z-Amine A™	10.0g
NaCl	5.0g
MgCl ₂ ·6H ₂ O	2.0g
Casamino acids solution	5.0mL

Casamino Acids Solution:

Composition per 10.0mL:

Casamino acids	2.0g

Preparation of Casamino Acids Solution: Add casamino acids to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except casamino acids solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 5.0mL of sterile casamino acids solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Escherichia coli*.

NZCYM Medium

Composition per liter:

Pancreatic digest of casein	10.0g
NaCl	5.0g
Yeast extract	5.0g
Maltose	2.0g
MgSO ₄ ·7H ₂ O	2.0g
Casamino acids	1.0g
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli* and *Pseudomonas* species.

NZY Agar**Composition per liter:**

Agar	20.0g
N-Z- Amine A.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
MgCl ₂ ·6H ₂ O	2.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation and maintenance of a variety of microorganisms.

NZY Agar**Composition per liter:**

Agar	11.0g
N-Z-Amine A.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
MgCl ₂ ·6H ₂ O	2.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and enumeration of a variety of microorganisms.

NZY Broth**Composition per liter:**

N-Z-Amine A.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
MgCl ₂ ·6H ₂ O	2.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of microorganisms.

O157:H7 ID Agar**Composition per liter:**

Proprietary

Source: This medium is available from bioMérieux

Use: A ready-to-use, chromogenic medium for strain-specific detection of *E. coli* O157:H7. A chromogenic medium for the detection of *Escherichia coli* O157:H7. In accordance with USDA, BAM and ISO 16654 standards, O157:H7 ID Agar allows the detection and presumptive identification of *E. coli* O157:H7.

Oatmeal Agar A

(ISP-3)

(DSMZ Medium 609)

Composition per liter:

Oatmeal (Quaker White Oats)	20.0g
Agar	18.0g
Trace salts solution	1.0mL
pH 7.0 ± 0.2 at 25°C	

Trace Salt Solution:**Composition per 100mL:**

FeSO ₄ ·7H ₂ O.....	0.1g
MnCl ₂ ·4H ₂ O	0.1g
ZnSO ₄ ·7H ₂ O	0.1g

Preparation of Trace Salt Solution: Add components to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add oats to 1.0L distilled/deionized water. Adjust pH to 7.2. Gently heat and bring to boiling. Cook or steam for 20 min. Filter through cheese cloth. Add 18.0g agar. Add 1.0mL trace salt solution. Bring volume to 1.0L distilled/deionized water. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into Petri dishes or distribute to sterile tubes.

Use: For the cultivation of various fungi.

Oatmeal Agar**Composition per 750.0mL:**

Agar	5.0g
Oatmeal, instant for babies	40.0g

Preparation of Medium: Add agar to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Add instant oatmeal for babies to distilled/deionized water and bring volume to 250.0mL. Mix thoroughly. Add the oatmeal solution to the melted agar solution. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation of fungi.

Oatmeal Agar (OA)

Composition per liter:

Agar	12.0g
Oatmeal, rolled oats	10.0g
Glycerin	5.0mL
Lactic acid	0.2mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of many filamentous fungi.

Oatmeal Salts Agar

Composition per liter:

Agar	18.0g
Oatmeal, baby (Beechnut™)	10.0g
MgSO ₄	1.0g
KH ₂ PO ₄	1.0g
NaNO ₃	1.0g

pH 5.6 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 5.6. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fungi.

Ogawa TB Medium

Composition per 300.0mL:

KH ₂ PO ₄	1.0g
Homogenized whole egg	200.0mL
Glycerol	6.0mL
Malachite Green (2% solution)	6.0mL

pH 6.5 ± 0.2 at 25°C

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18–24

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Add components, except homogenized whole egg, to distilled/deionized

water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 200.0mL of sterile homogenized whole egg. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes in 7.0mL volumes. Inspissate at 85°–90°C (moist heat) for 60 min.

Use: For the isolation and cultivation of *Mycobacterium* species, except for *Mycobacterium leprae*.

Oleic Albumin Complex

Composition per liter:

Bovine albumin fraction V	50.0g
NaCl	8.5g
Oleic acid	0.6mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: For use in media employed for the cultivation of mycobacteria.

ONPG Broth

Composition per liter:

Peptone water	750.0mL
ONPG solution	250.0mL

pH 7.2–7.4 at 25°C

ONPG Solution:

Composition per 250.0mL:

ONPG (<i>o</i> -Nitrophenyl- β - D-galactopyranoside)	1.5g
Sodium phosphate buffer (0.01M, pH 7.5)	250.0mL

Preparation of ONPG Solution: Add ONPG to 250.0mL of sodium phosphate buffer. Mix thoroughly. Filter sterilize.

Peptone Water:

Composition per 750.0mL:

Peptone	7.5g
NaCl	3.75g

Preparation of Peptone Water: Add components to distilled/deionized water and bring volume to 750.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 8.0–8.4. Continue boiling for 10 min. Filter through Whatman #1 filter paper. Readjust pH of filtrate to 7.2–7.4. Autoclave for 20 min at 10 psi pressure–115°C. Cool to 25°C.

Preparation of Medium: Aseptically combine the sterile ONPG solution with the cooled, sterile peptone water. Mix thoroughly. Aseptically distribute into tubes in 2.5–3.0.0mL volumes. Store at 4°C for up to 1 month.

Use: For the differentiation of a variety of Gram-negative bacteria based on production of β -galactosidase. For the differentiation of lactose-delayed bacteria from lactose-negative bacteria. For the differentiation of *Pseudomonas cepacia* (positive) and *Pseudomonas maltophilia* (positive) from other *Pseudomonas* species (negative). Bacteria that produce β -galactosidase turn the medium yellow.

OR Indicator Agar (Oxidation-Reduction Indicator Agar)

Composition per liter:

Agar	15.0g
Sodium glycerol phosphate.....	10.0g
Sodium thioglycolate	1.7g
CaCl ₂ ·2H ₂ O.....	0.1g
Methylene Blue	6.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For use as an indicator of oxygen-free conditions in anaerobic culture chambers.

Orange Serum Agar

Composition per liter:

Agar	17.0g
Pancreatic digest of casein.....	10.0g
Glucose	4.0g
Yeast extract.....	3.0g
K ₂ HPO ₄	2.5g
Orange serum.....	200.0mL
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of microorganisms associated with the spoilage of citrus products. For the cultivation of lactobacilli, other aciduric microorganisms, and pathogenic fungi.

Orange Serum Agar

Composition per liter:

Agar	14.0g

Pancreatic digest of casein.....	10.0g
Glucose	4.0g
Orange serum, equivalent solids.....	3.5g
Yeast extract.....	3.0g
K ₂ HPO ₄	2.5g
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and enumeration of spoilage organisms of citrus products. For the enumeration and cultivation of microorganisms from citrus juice and other products. For the cultivation of lactobacilli, pathogenic fungi, and other aciduric microorganisms.

Orange Serum Agar

Composition per liter:

Agar	15.5g
Orange serum.....	10.0g
Pancreatic digest of casein.....	10.0g
Glucose	4.0g
Yeast extract.....	3.0g
K ₂ HPO ₄	2.5g
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration and cultivation of microorganisms from citrus juice and other products. For the cultivation of lactobacilli, pathogenic fungi, and other aciduric microorganisms.

Orange Serum Broth Concentrate 10X

Composition per liter:

Pancreatic digest of casein.....	10.0g
Glucose	4.0g
Yeast extract.....	3.0g
K ₂ HPO ₄	2.5g
Orange serum concentrate	100.0mL
pH 5.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed solution from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and enumeration of microorganisms associated with the spoilage of citrus products. For the cultivation of lactobacilli, other aciduric microorganisms, and pathogenic fungi.

Organic Acid Medium KP (Organic Acid Medium, Kauffmann and Petersen)

Composition per liter:

Gelatin.....	10.0g
Bromthymol Blue	0.024g
Organic acid solution	100.0mL
pH 7.4 ± 0.2 at 25°C	

Organic Acid Solution:

Composition per 100.0mL:

Organic acid	10.0g

Preparation of Medium: Add organic acid to distilled/deionized water and bring volume to 100.0mL. Sodium potassium D-tartrate, sodium citrate, or mucic acid may be used. Mix thoroughly.

Preparation of Medium: Add components, except organic acid solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile organic acid solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and differentiation of members of the *Enterobacteriaceae* based on their ability to utilize different organic acids as carbon source. Bacteria that utilize tartrate, citrate, or mucate turn the medium yellow.

Ornithine Broth (BAM M44)

Composition per liter:

L-Ornithine.....	5.0g
Peptone or gelysate	5.0g
Yeast extract.....	3.0g
Glucose	1.0g
Bromcresol Purple	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after

sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure–121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of bacteria based on their ability to decarboxylate the amino acid ornithine. Bacteria that decarboxylate ornithine turn the medium turbid purple.

Ornithine Broth with NaCl (BAM M44)

Composition per liter:

L-Ornithine.....	5.0g
Peptone or gelysate	5.0g
Yeast extract.....	3.0g
Glucose	1.0g
Bromcresol Purple	0.02g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH so that it will be 6.5 ± 0.2 after sterilization. Distribute into 16 × 150mm screw-capped tubes in 5.0mL volumes. Autoclave medium with loosely capped tubes for 10 min at 15 psi pressure–121°C. Screw the caps on tightly for storage and after inoculation.

Use: For the cultivation and differentiation of *Vibrio* spp. based on their ability to decarboxylate the amino acid ornithine. Bacteria that decarboxylate ornithine turn the medium turbid purple.

Oxford Agar (Listeria Selective Agar, Oxford)

Composition per liter:

Special peptone.....	23.0g
LiCl.....	15.0g
Agar	10.0g
NaCl.....	5.0g
Cornstarch.....	1.0g
Esculin	1.0g
Ferric ammonium citrate.....	0.5g
Antibiotic inhibitor	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Antibiotic Inhibitor:

Composition per 10.0mL:

Cycloheximide.....	0.4g
Colistin sulfate.....	0.02g
Fosfomycin.....	0.01g
Acriflavine	5.0mg

Cefotetan	2.0mg
Ethanol (50% solution)	10.0mL

Preparation of Antibiotic Inhibitor: Add antibiotics to 10.0mL of ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic inhibitor, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes* from specimens containing a mixed bacterial flora.

Oxford Agar, Modified
(*Listeria* Selective Agar,
Modified Oxford)
(MOX Agar)

Composition per liter:

Special peptone	23.0g
LiCl	15.0g
Agar	12.0g
NaCl	5.0g
Cornstarch	1.0g
Esculin	1.0g
Ferric ammonium citrate	0.5g
Antibiotic inhibitor	10.0mL
pH 7.0 ± 0.2 at 25°C	

Antibiotic Inhibitor:

Composition per 10.0mL:

Moxalactam	0.015g
Colistin sulfate	0.01g

Preparation of Antibiotic Inhibitor: Add components to distilled /deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic inhibitor, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 10 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes* from specimens containing a mixed bacterial flora.

Oxford Medium
(BAM M118)

Composition per liter:

Special peptone	23.0g
LiCl	15.0g
Agar	10.0g
NaCl	5.0g
Cornstarch	1.0g
Esculin	1.0g
Ferric ammonium citrate	0.5g
Antibiotic inhibitor	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Supplement Mix:

Composition per 10.0mL:

Cycloheximide	0.4g
Colistin sulfate	0.02g
Fosfomycin	0.01g
Acriflavine	5.0mg
Cefotetan	2.0mg
Ethanol (50% solution)	10.0mL

Preparation of Supplement Mix: Add components to 10.0mL of ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except supplement mix, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL of sterile supplement mix. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes* from specimens containing a mixed bacterial flora.

Oxidative-Fermentative
Glucose Medium, Semisolid
(OF Glucose Medium, Semisolid)
(BAM M116)

Composition per liter:

Glucose	10.0g
NaCl	5.0g
Agar	2.0g
Pancreatic digest of casein	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue dye	0.08g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For differentiating Gram-negative bacteria based upon determining the oxidative and fermentative metabolism of glucose. Bacteria that ferment glucose turn the medium yellow. For *Campylobacter* detection prepare half of the tubes with glucose and half without.

**Oxidative-Fermentative
Glucose Medium, Semisolid with NaCl
(OF Glucose Medium,
Semisolid with NaCl)
(BAM M116)**

Composition per liter:

NaCl.....	20.0g
Glucose	10.0g
Agar	2.0g
Pancreatic digest of casein.....	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue dye.....	0.08g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For differentiating Gram-negative halophilic *Vibrio* spp. based upon determining the oxidative and fermentative metabolism of glucose. Bacteria that ferment glucose turn the medium yellow. For *Campylobacter* detection prepare half of the tubes with glucose and half without.

**Oxidation-Fermentation Medium
(OF Medium)**

Composition per liter:

NaCl.....	5.0g
Agar	2.5g
Pancreatic digest of casein.....	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue	0.03g
Carbohydrate solution.....	100.0mL

pH 6.8 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For differentiating Gram-negative bacteria based upon determining the oxidative and fermentative metabolism of carbohydrates.

**Oxidation-Fermentation
Medium, Hugh-Leifson's
(Hugh-Leifson's Oxidation
Fermentation Medium)**

Composition per liter:

NaCl.....	5.0g
Agar	3.0g
Peptone	2.0g
K ₂ HPO ₄	0.3g
Carbohydrate solution.....	100.0mL
Bromthymol Blue solution (0.2%)	15.0mL

pH 7.1 ± 0.2 at 25°C

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For differentiating Gram-negative bacteria, such as *Vibrio* species, based upon determining the oxidative and fermentative metabolism of carbohydrates. Bacteria that ferment the carbohydrate turn the medium yellow.

**Oxidation-Fermentation Medium, King's
(King's OF Medium)**

Composition per liter:

Base:

Agar	3.0g

Pancreatic digest of casein.....	2.0g
Carbohydrate solution.....	100.0mL
Phenol Red (1.5% solution).....	2.0mL
pH to 7.3 ± 0.2	

Carbohydrate Solution:**Composition per 100.0mL:**

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For differentiating bacteria based upon determining the oxidative and fermentative metabolism of carbohydrates. Bacteria that ferment the carbohydrate turn the medium yellow.

**Oxidative-Fermentative
Glucose Medium, Semisolid, with NaCl
(OF Glucose Medium, Semisolid
with NaCl)**

Composition per liter:

NaCl.....	20.0g
Glucose	10.0g
Agar	2.0g
Pancreatic digest of casein.....	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue dye.....	0.08g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For differentiating halophilic *Vibrio* species based upon determining the oxidative and fermentative metabolism of glucose. Bacteria that ferment glucose turn the medium yellow.

**Oxidative-Fermentative Medium
(OF Medium)**

Composition per liter:

NaCl.....	5.0g
Agar	2.0g

Pancreatic digest of casein.....	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue	0.08g
Carbohydrate solution.....	100.0mL
pH 6.8 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Carbohydrate Solution:**Composition per 100.0mL:**

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbohydrate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For differentiating bacteria based upon determining the oxidative and fermentative metabolism of carbohydrates. Bacteria that ferment the carbohydrate turn the medium yellow.

**Oxidative-Fermentative Test Medium
(OF Test Medium)
(BAM M117)**

Composition per liter:

NaCl.....	5.0g
Agar	2.5g
Peptone	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue	0.03g
Carbohydrate solution.....	100.0mL

Carbohydrate Solution:**Composition per 100.0mL:**

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 3.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.3mL of sterile carbohydrate solution to each tube. Mix thoroughly.

Use: For the cultivation and differentiation of a variety of microorganisms based on their ability to ferment a specific carbohydrate. Bacteria that ferment the specific carbohydrate turn the medium yellow.

**Oxidative-Fermentative Test Medium
with NaCl
(OF Test Medium with NaCl)
(BAM M117)**

Composition per liter:

NaCl.....	5.0g
Agar	2.5g
Peptone.....	2.0g
K ₂ HPO ₄	0.3g
Bromthymol Blue	0.03g
Carbohydrate solution.....	100.0mL

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 3.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.3mL of sterile carbohydrate solution to each tube. Mix thoroughly.

Use: For the cultivation and differentiation of a halophilic *Vibrio* spp. based on their ability to ferment a specific carbohydrate. Bacteria that ferment the specific carbohydrate turn the medium yellow.

**Oxytetracycline Glucose
Yeast Extract Agar
(OGYE Agar)**

Composition per liter:

Glucose	20.0g
Agar	12.0g
Yeast extract.....	5.0g
Biotin	0.1mg
Oxytetracycline solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Oxytetracycline Solution:

Composition per 10.0mL:

Oxytetracycline.....	0.1g

Tris(hydroxymethyl) aminomethane

buffer (0.1M, pH 7.0)	10.0mL

Preparation of Oxytetracycline Solution:

Add oxytetracycline to 10.0mL of Tris buffer. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except oxytetracycline solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 10 min at 10 psi pressure–115°C. Cool to 45°–50°C. Aseptically add sterile oxytetracycline solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, enumeration, and cultivation of yeasts and other fungi from foods.

**Oxytetracycline Glucose
Yeast Extract Agar
(OGYE Agar)**

Composition per liter:

Glucose	20.0g
Agar	12.0g
Yeast extract.....	5.0g
Oxytetracycline solution.....	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Oxytetracycline Solution:

Composition per 10.0mL:

Oxytetracycline.....	0.1g

Preparation of Oxytetracycline Solution:

Add oxytetracycline to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except oxytetracycline solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile oxytetracycline solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, enumeration, and cultivation of yeasts and other fungi from foods.

OZR Medium

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	1.0g
Yeast extract.....	1.0g

Preparation of Medium: Add components to seawater and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Leucothrix mucor*.

P Agar

Composition per liter:

Agar	15.0g
Peptone.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Glucose	1.0g

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Staphylococcus* species.

PA-C Agar (mPA-C Agar)

Composition per liter:

Agar	12.0g
L-Lysine-HCl.....	5.0g
NaCl.....	5.0g
Na ₂ S ₂ O ₃	5.0g
Yeast extract.....	2.0g
MgSO ₄ ·7H ₂ O	1.5g
Lactose	1.25g
Sucrose.....	1.25g
Xylose	1.25g
Ferric ammonium citrate.....	0.8g
Phenol Red.....	0.08g
Nalidixic acid.....	0.037g
Kanamycin	8.0mg

pH 7.2 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective recovery and enumeration of *Pseudomonas aeruginosa* from water samples.

Pablum Cereal Agar

Composition per liter:

Pablum cereal, precooked.....	100.0g
Agar	18.0g
Chloramphenicol.....	0.05g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of dematiaceous fungi and stimulation of spore formation.

Pai Medium

Composition per liter:

Homogenized whole egg	666.0mL
NaCl (0.85% solution)	334.0mL

pH 6.75 ± 0.2 at 25°C

Homogenized Whole Egg:

Composition per liter:

Whole eggs	18-24

Preparation of Homogenized Whole Egg: Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 1.0L.

Preparation of Medium: Combine components. Mix thoroughly. Aseptically distribute into sterile tubes. Inspissate tubes in a slanted position at 80–90°C (moist heat) for 30 min.

Use: For the maintenance of stock cultures of *Salmonella typhi* and other *Salmonella* species.

PALCAM Agar (Polymyxin Acriflavine LiCl Ceftazidime Esculin Mannitol Agar) (PALCAM *Listeria* Selective Agar) (BAM M118a)

Composition per liter:

Peptone	23.0g
LiCl.....	15.0g
Agar	10.0g
Mannitol.....	10.0g
NaCl.....	5.0g

Yeast extract.....	3.0g
Starch	1.0g
Esculin	0.8g
Ferric ammonium citrate.....	0.5g
Glucose	0.5g
Phenol Red.....	0.08g
PALCAM selective supplement.....	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

PALCAM Selective Supplement:

Composition per 10.0mL:

Ceftazidime.....	20.0mg
Polymyxin B	10.0mg
Acridiflavine·HCl.....	5.0mg

Preparation of PALCAM Selective Supplement: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Egg Yolk Emulsion:

Composition:

Chicken egg yolks.....	11
Whole chicken egg.....	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Preparation of Medium: Add components, except PALCAM selective supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile PALCAM selective supplement. Mix thoroughly. The addition of 25.0mL of egg yolk emulsion may aid in the recovery of damaged *Listeria*. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and differentiation of *Listeria monocytogenes* and other *Listeria* species from foods.

Papain (BAM M56a)

Composition per 100mL:

Papain.....	5.0g

Preparation of Medium: Add 5.0g papain to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Use: As a replacement for gelatinase.

Park and Sanders Enrichment Broth

Composition per 1010.0mL:

Basal medium	1.0L
Supplement A	5.0mL
Supplement B.....	5.0mL
pH 7.0 ± 0.2 at 25°C	

Basal Medium:

Composition per liter:

Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Glucose	1.0g
Sodium pyruvate.....	0.25g
NaHSO ₃	0.1g
Horse blood, lysed	50.0mL

Preparation of Basal Medium: Add components, except horse blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add sterile horse blood. Mix thoroughly.

Supplement A:

Composition per 5.0mL:

Vancomycin	0.01g
Trimethoprim lactate.....	0.01g

Preparation of Supplement A: Add components to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Supplement B:

Composition per 5.0mL:

Cefoperazone	0.032g
Cycloheximide.....	0.1g

Preparation of Supplement B: Add components to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 1.0L of cooled, sterile basal medium, aseptically add 5.0mL of sterile supplement A. Mix thoroughly. Aseptically distribute into flasks in 100.0mL volumes. Inoculate medium with food samples. Incubate at 31°–32°C for 4 hr to recover and resuscitate injured cells. Aseptically add 0.5mL of supplement B to each 100.0mL of medium. Incubate cultures at 37°C for 2 hr.

Use: For the cultivation and enrichment of *Campylobacter* species from foods.

PE2 Medium

Composition per liter:

Peptone	20.0g

Yeast extract.....	3.0g
Alaska seed peas.....	416–520
Bromcresol Purple solution	2.0mL

Bromcresol Purple Solution:**Composition per 100.0mL:**

Bromcresol Purple	2.0g
Ethanol	10.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to 10.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Filter sterilize.

Preparation of Medium: Add components, except Alaska seed peas, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Add 8–10 Alaska seed peas to each of 18 × 150 mm screw-capped tubes. Distribute the broth into each tube in 19.0mL volumes. Allow tubes to stand for 1 hr. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Clostridium botulinum* from foods.

Pectin Medium**Composition per liter:**

Pectin	30.0g
Yeast extract.....	5.0g
Bromthymol Blue (0.1% solution).....	1.0mL
CaCl ₂ ·2H ₂ O (10.0% solution).....	0.6mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add 100.0mL of distilled/deionized water to a 2.0L flask and place on a magnetic stirrer with no heat. While stirring, slowly add CaCl₂·2H₂O, Bromthymol Blue solution, yeast extract, and pectin. Add slowly to ensure each particle is wetted. Gently heat and bring to almost boiling. Adjust pH to 7.3 with 1N NaOH. Do not overshoot pH 7.3.

Use: For the isolation and presumptive identification of *Yersinia enterocolitica* and *Yersinia pseudotuberculosis* from other fermenting Gram-negative bacilli such as *Enterocolitica agglomerans* which is often confused with *Yersinia*. *Enterocolitica agglomerans* does not produce pectinase. *Yersinia enterocolitica* is strongly positive for pectinase activity. *Yersinia pseudotuberculosis* is weakly positive for pectinase activity. *Yersinia pestis* is negative for pectinase activity. Also used for the differentiation of *Klebsiella oxytoca*, which is pectinase positive.

**Penicillin-Streptomycin Solution
(BAM M119)****Composition per 100.0mL:**

Penicillin G.....	500,000IU
Streptomycin.....	500,000IU

Preparation of Penicillin-Streptomycin Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Store at 5°C.

Use: An antibiotic concentrate used as a selective supplement for various media, e.g. 10.0mL per liter of penicillin-streptomycin solution is used in media for the isolation of *Vibrio cholerae* from water and foods.

Peptone Iron Agar**Composition per liter:**

Agar	15.0g
Peptone	15.0g
Proteose peptone.....	5.0g
Sodium glycerophosphate.....	1.0g
Ferric ammonium citrate.....	0.5g
Na ₂ S ₂ O ₃	0.08g

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the cultivation and differentiation of microorganisms based on their ability to produce H₂S. Microorganisms that produce H₂S turn the medium black.

Peptone Medium**Composition per liter:**

Peptone.....	10.0g

Preparation of Medium: Add peptone to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli*.

**Peptone Sorbitol Bile Broth
(BAM M120)****Composition per liter:**

Sorbitol	10.0g
Na ₂ HPO ₄	8.23g
NaCl.....	5.0g

Peptone.....	5.0g
Bile salts No. 3.....	1.5g
NaH ₂ PO ₄	1.2g

pH 7.6 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into bottles in 100.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enrichment and cultivation of *Yersinia* species.

Peptone Water

Composition per liter:

Peptone.....	10.0g
NaCl.....	5.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of nonfastidious microorganisms, for carbohydrate fermentation tests, and for performing the indole test.

Peptone Water with Andrade's Indicator

Composition per liter:

Peptone.....	10.0g
NaCl.....	5.0g
Andrade's indicator.....	100.0mL
Carbohydrate solution.....	20.0mL

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Andrade's Indicator:

Composition per 100.0mL:

Acid Fuchsin.....	0.1g
NaOH (1N solution).....	16.0mL

Caution: Acid Fuchsin is a potential carcinogen and care must be taken to avoid inhalation of the powdered dye and contact with the skin.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 9.8mL of medium. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube.

Use: For use in carbohydrate fermentation tests. Fermentation is determined by the production of acid—broth turns pink—and formation of gas—bubble trapped in Durham tube.

Peptone Yeast Trypticase Agar (ATCC Medium 118)

Composition per liter:

Agar.....	15.0g
Peptone.....	6.0g
Trypticase (pancreatic digest of casein).....	4.0g
Yeast extract.....	3.0g
Beef extract.....	1.5g
Glucose.....	1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of heterotrophic bacteria.

Pfizer Selective *Enterococcus* Agar (PSE Agar)

Peptone C.....	17.0g
Agar.....	15.0g
Bile.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Peptone B.....	3.0g
Esculin.....	1.0g
Sodium citrate.....	1.0g
Ferric ammonium citrate.....	0.5g
NaN ₃	0.25g

pH 7.1 ± 0.2 at 25°C

Caution: Sodium azide is toxic. Azides also react with metals and disposal must be highly diluted.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and enumeration of *Enterococcus* species by the multiple tube technique.

PGA
(Potato Glucose Agar)

Composition per liter:

Potatoes	500.0g
Glucose	20.0g
Agar	15.0g
Yeast extract	5.0g

Preparation of Medium: Slice potatoes with skin. Place 500.0g of potatoes in 1.0L of distilled/deionized water. Gently heat and bring to boiling. Allow to boil for 20 min. Filter through Whatman filter paper. Add agar and other components to filtrate. Bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of numerous filamentous fungi.

PGLE Medium
(Peptone Glucose
Liver Extract Medium)

Composition per liter:

Agar	25.0g
Glucose	20.0g
Peptone.....	5.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	1.0g
Liver extract.....	0.5g

pH 9.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Enterobacter cloacae*.

Phenethyl Alcohol Agar
(Phenylethanol Agar)
(Phenethyl Alcohol Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	15.0g

NaCl.....	5.0g
Papaic digest of soybean meal.....	5.0g
β-Phenethyl alcohol.....	2.5g
Blood.....	50.0mL

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add sterile defibrinated blood. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of Gram-positive bacteria, particularly Gram-positive cocci, from specimens with a mixed flora. Do not use for the observation of hemolytic reactions.

Phenol Red Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
Phenol Red.....	0.018g
Carbohydrate solution.....	20.0mL

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 20.0mL of sterile carbohydrate solution. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the determination of fermentation reactions. Bacteria that can ferment the added carbohydrate turn the medium yellow.

Phenol Red Agar

Composition per liter:

Agar	15.0g
Proteose peptone No. 3	10.0g

NaCl.....	5.0g
Beef extract.....	1.0g
Phenol Red.....	0.025g
Carbohydrate solution.....	20.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 20.0mL of sterile carbohydrate solution. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the determination of fermentation reactions. Bacteria that can ferment the added carbohydrate turn the medium yellow.

Phenol Red Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
Phenol Red.....	0.018g
Carbohydrate solution.....	20.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 9.8mL of medium. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube.

Use: For the determination of fermentation reactions in the differentiation of microorganisms. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Carbohydrate Broth (BAM M121)

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Carbohydrate solution.....	200.0mL
Phenol Red solution.....	7.2mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostics.

Phenol Red Solution:

Composition per 10.0mL:

Phenol Red.....	0.025g

Preparation of Phenol Red Solution: Add Phenol Red to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Carbohydrate Solution:

Composition per 200.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate (5.0g dulcitol, 10.0g lactose, or 10.0g sucrose) to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 8.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add 2.0mL of sterile carbohydrate solution to each tube.

Use: For the determination of fermentation reactions in the differentiation of microorganisms, such as *Salmonella* spp. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Carbohydrate Broth with NaCl (BAM M121)

Composition per liter:

NaCl.....	30.0g
Pancreatic digest of casein.....	10.0g
Beef extract.....	1.0g

Carbohydrate solution.....	200.0mL
Phenol Red solution.....	7.2mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium without added NaCl is available as a premixed powder from BD Diagnostics.

Phenol Red Solution:

Composition per 10.0mL:

Phenol Red.....	0.025g

Preparation of Phenol Red Solution: Add Phenol Red to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Carbohydrate Solution:

Composition per 200.0mL:

Carbohydrate.....	5.0–10.0g

Preparation of Carbohydrate Solution: Add carbohydrate (5.0g ducitol, 10.0g lactose, or 10.0g sucrose) to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 8.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add 2.0mL of sterile carbohydrate solution to each tube.

Use: For the determination of fermentation reactions in the differentiation of halophilic *Vibrio* spp. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Glucose Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
Glucose.....	5.0g
NaCl.....	5.0g
Phenol Red.....	0.018g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.3 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C.

Use: For determination of the ability of a microorganism to ferment glucose. Fermentation is determined by the production of acid—broth turns

yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Glucose Broth (BAM M122)

Composition per liter:

Proteose peptone No. 3.....	10.0g
Glucose.....	5.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Phenol Red solution.....	7.2mL

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostics.

Phenol Red Solution:

Composition per 10.0mL:

Phenol Red.....	0.025g

Preparation of Phenol Red Solution: Add Phenol Red to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 10 min at 13 psi pressure–118°C.

Use: For determination of the ability of a microorganism to ferment glucose. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Glucose Broth with NaCl (BAM M122)

Composition per liter:

NaCl.....	30.0g
Proteose peptone No. 3.....	10.0g
Glucose.....	5.0g
Beef extract.....	1.0g
Phenol Red solution.....	7.2mL

pH 7.4 ± 0.2 at 25°C

Source: This medium without added NaCl is available as a premixed powder from BD Diagnostics.

Phenol Red Solution:

Composition per 10.0mL:

Phenol Red.....	0.025g

Preparation of Phenol Red Solution: Add Phenol Red to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 10 min at 13 psi pressure–118°C.

Use: For determination of the ability of halophilic *Vibrio* spp. to ferment glucose. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Lactose Agar

Composition per liter:

Agar	15.0g
Lactose	10.0g
Proteose peptone No. 3	10.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Phenol Red.....	25.0mg
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For determination of the ability of a microorganism to ferment lactose. Fermentation is determined by the production of acid—medium turns yellow.

Phenol Red Lactose Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
Lactose	5.0g
NaCl.....	5.0g
Phenol Red.....	0.018g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.3 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C.

Use: For determination of the ability of a microorganism to ferment lactose. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Mannitol Agar

Composition per liter:

Agar	15.0g
Mannitol.....	10.0g
Proteose peptone No. 3	10.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Phenol Red.....	25.0mg

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For determination of the ability of a microorganism to ferment mannitol. Fermentation is determined by the production of acid—medium turns yellow.

Phenol Red Mannitol Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
D-Mannitol.....	5.0g
NaCl.....	5.0g
Phenol Red.....	0.018g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.3 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C.

Use: For determination of the ability of a microorganism to ferment mannitol. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Sucrose Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g

Sucrose.....	5.0g
Phenol Red.....	0.018g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.3 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 10.0mL of medium. Autoclave for 15 min at 13 psi pressure–118°C.

Use: For determination of the ability of a microorganism to ferment sucrose. Fermentation is determined by the production of acid—broth turns yellow—and formation of gas—bubble trapped in Durham tube.

Phenol Red Tartrate Agar

Composition per liter:

Agar	15.0g
Peptone.....	10.0g
Potassium tartrate.....	10.0g
NaCl.....	5.0g
Phenol Red.....	0.024g
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to cold distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in an upright position.

Use: For the differentiation of Gram-negative bacteria of the intestinal groups, particularly members of the *Salmonella* (paratyphoid) group based on their ability to ferment tartrate.

Phenol Red Tartrate Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
Potassium tartrate.....	10.0g
Agar	5.0g
NaCl.....	5.0g
Phenol Red.....	0.024g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of Gram-negative bacteria of the intestinal groups, particularly members of the *Salmonella* (paratyphoid) group based on their ability to ferment tartrate.

Phenylalanine Agar (Phenylalanine Deaminase Medium) (BAM M123)

Composition per liter:

Agar	12.0g
NaCl.....	5.0g
Yeast extract.....	3.0g
DL-Phenylalanine	2.0g
Na ₂ HPO ₄	1.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of enteric Gram-negative bacilli on the basis of their ability to produce phenylpyruvic acid from phenylalanine. After appropriate incubation of bacteria, ferric chloride reagent is added on the agar. Formation of a green color in 1–5 min indicates the production of phenylpyruvic acid.

Phenylalanine Agar (Phenylalanine Deaminase Medium) (BAM M123)

Composition per liter:

Agar	12.0g
NaCl.....	5.0g
Yeast extract.....	3.0g
L-Phenylalanine	1.0g
Na ₂ HPO ₄	1.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of enteric Gram-negative bacilli on the basis of their ability to produce phenylpyruvic acid from phenylalanine. After appro-

prate incubation of bacteria, ferric chloride reagent is added on the agar. Formation of a green color in 1–5 min indicates the production of phenylpyruvic acid.

Phenylalanine Malonate Broth

Composition per liter:

Sodium malonate	3.0g
DL-Phenylalanine	2.0g
NaCl	2.0g
(NH ₄) ₂ SO ₄	2.0g
Yeast extract	1.0g
K ₂ HPO ₄	0.6g
KH ₂ PO ₄	0.4g
Bromthymol Blue	0.025g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–115°C.

Use: For the differentiation of Gram-negative enteric bacilli on the basis of malonate utilization and formation of pyruvic acid from phenylalanine.

Phenylethanol Agar

Composition per liter:

Agar	15.0g
Tryptose	10.0g
NaCl	5.0g
Beef extract	3.0g
Phenylethanol	2.5g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation of staphylococci and streptococci from specimens containing a mixed flora.

Phenylethanol Blood Agar

Composition per liter:

Agar	15.0g
Tryptose	10.0g
NaCl	5.0g

Beef extract	3.0g
Phenylethanol	2.5g
Blood, defibrinated	50.0mL

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components, except blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Aseptically add sterile defibrinated blood. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation of staphylococci and streptococci from specimens containing a mixed flora.

Pisu Medium

Composition per 1512.0mL:

Agar base	960.0mL
Horse serum, sterile	360.0mL
L-Cystine solution	180.0mL
Lead acetate solution	12.0mL

pH 6.8 ± 0.2 at 25°C

Agar Base:

Composition per liter:

Proteose peptone No. 3	20.0g
Agar	7.0g
NaCl	5.0g
Meat extract	4.0g

Preparation for Agar Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Dissolve in steam for 15 min at 0 psi pressure–100°C. Cool to 45°–50°C. Adjust pH to 7.5. Filter sterilize. Distribute into 200.0mL Erlenmeyer flasks in 80.0mL volumes. Autoclave for 60 min at 0 psi pressure–100°C. Cool to 60°C.

L-Cystine Solution:

Composition per 200.0mL:

L-Cystine	2.0g

Preparation of L-Cystine Solution: Add L-cystine to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Filter sterilize.

Lead Acetate Solution:

Composition per 100.0mL:

Lead acetate	10.0g

Preparation of Lead Acetate Solution: Add lead acetate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation for Medium: To each flask containing 80.0mL of cooled, sterile agar base, aseptically add 30.0mL of sterile horse serum, 15.0mL of sterile L-cystine solution, and 1.0mL of sterile lead acetate

solution. Mix thoroughly. Aseptically distribute into small sterile tubes in 2.0–3.0mL volumes.

Use: For the cultivation and differentiation of bacteria based on their ability to produce cystinase. Cystinase-producing bacteria turn the medium black.

PL Agar

Composition per liter:

Agar	15.0g
Mannitol.....	7.5g
L-Arabinose.....	5.0g
Peptone.....	5.0g
NaCl.....	5.0g
Lysine.....	2.0g
Yeast extract.....	2.0g
Bile salts No. 2.....	1.0g
Inositol	1.0g
Phenol Red.....	0.08g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.4. Distribute into tubes or flasks. Autoclave for 15 min at 10 psi pressure–115°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Pleisomonas shigelloides* from foods.

Plate Count Agar (ATCC Medium 1048) (BAM M124)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	2.5g
Glucose	1.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of bacteria in milk, water, food, and dairy products.

Plate Count Agar, Modified

Composition per liter:

Pancreatic digest of casein.....	20.0g
Yeast extract.....	20.0g
Agar	10.0g
Glucose	4.0g

pH 7.0 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into bottles. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and enumeration of microorganisms from food by the plate count method.

Plate Count Agar, Special

Composition per liter:

Agar	30.1g
Pancreatic digest of casein.....	6.13g
Yeast extract.....	3.06g
Glucose	1.23g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of viable bacteria in raw milk, milk, and other dairy products.

Plate Count Agar with Antibiotic

Composition per liter:

Agar	9.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	2.5g
Glucose	1.0g
Chloramphenicol.....	0.1g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts from foods.

Plate Count Agar with Antibiotic-Free Skim Milk

Composition per liter:

Agar	10.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	2.5g
Skim milk, antibiotic free	1.0g
Glucose	1.0g
pH 6.9 $\pm$ 0.1 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of viable bacteria in milk and dairy products.

Plate Count Broth (m-Plate Count Broth)

Composition per liter:

Yeast extract.....	5.0g
Glucose	2.0g
pH 7.0 $\pm$ 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the determination of bacterial counts by the membrane filter method.

PM Indicator Agar

Composition per liter:

Agar	15.0g
Glucose	5.25g
Peptone.....	5.0g
Beef extract.....	3.0g
Pancreatic digest of casein.....	1.7g
Sorbitan monooleate complex	1.0g
NaCl.....	0.5g
Papaic digest of soybean meal.....	0.3g
K ₂ HPO ₄	0.25g
Bromocresol Purple	0.06g
pH 7.8 $\pm$ 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Inoculate medium with a suspension of *Bacillus stearothermophilus* spores. Pour into 100mm flat-bottomed Petri dishes in 6.0mL volumes. Use medium immediately.

Use: For the rapid detection of trace amounts of penicillin in milk using the AOAC *Bacillus stearothermophilus* Qualitative Disc Method II.

PMP Broth (BAM M125)

Composition per liter:

Na ₂ HPO ₄	7.9g
D-Mannitol	2.5g
Peptone	2.5g
NaH ₂ PO ₄	1.1g
pH 7.6 $\pm$ 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enrichment and cultivation of *Yersinia pseudotuberculosis* from food.

Poly- β -Hydroxybutyrate Medium (PHB Medium)

Composition per liter:

Part A	900.0mL
Part B	100.0mL
pH 7.2 $\pm$ 0.2 at 25°C	

Part A:

Composition per 900.0mL:

K ₂ HPO ₄ ·3H ₂ O	0.6g
KH ₂ PO ₄	0.2g
MgSO ₄ ·7H ₂ O	0.2g
(NH ₄) ₂ SO ₄	0.2g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.2. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Part B:

Composition per 100.0mL:

Glucose	10.0g

Preparation of Medium: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Preparation of Medium: Aseptically combine 900.0mL of cooled, sterile part A and 100.0mL of cooled, sterile part B. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and differentiation of *Pseudomonas* species based on their ability to produce intracellular poly- β -hydroxybutyrate. Production of poly- β -hydroxybutyrate is determined by staining cells with Sudan Black B.

**Polymyxin Pyruvate Egg Yolk Mannitol
Bromthymol Blue Agar
(PEMBA)**

Composition per 110.0mL:

Agar	1.8g
D-Mannitol	1.0g
Sodium pyruvate	1.0g
Na ₂ HPO ₄	0.25g
NaCl	0.2g
Peptone	0.1g
KH ₂ PO ₄	0.025g
Bromthymol Blue	0.01g
MgSO ₄ ·7H ₂ O	0.01g
Antibiotic inhibitor	5.0mL
Egg yolk emulsion (20% solution)	5.0mL
pH 7.4 ± 0.2 at 25°C	

Antibiotic Inhibitor:

Composition per 5.0mL:

Polymyxin B	10,000U

Preparation of Antibiotic Inhibitor: Add components to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Egg Yolk Emulsion, 20% Solution:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	80.0mL

Preparation of Egg Yolk Emulsion, 20% Solution: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 20.0mL of egg yolk emulsion and add to 80.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Preparation of Medium: Add components, except antibiotic inhibitor and egg yolk emulsion, 20%, to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic inhibitor and egg yolk emulsion, 20%. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of *Bacillus cereus*.

Polymyxin *Staphylococcus* Medium

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
Lecithin	0.7g
Polymyxin	0.075g
Tween 80	10.2mL
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the selective isolation and cultivation of pathogenic, coagulase-positive *Staphylococcus aureus*. *Proteus* species will grow on this medium but appear as translucent colonies.

Polypectate Gel Medium

Composition per liter:

Sodium polypectate	70.0g
K ₂ HPO ₄	5.0g
Peptone	5.0g
KH ₂ PO ₄	1.0g
CaCl ₂ ·2H ₂ O	0.6g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sodium polypectate, to 500.0mL of boiling water. Put into a blender and mix on low speed to dissolve. Add the sodium polypectate slowly with the blender on low speed to minimize air bubbles. Adjust pH to 7.0. Bring volume to 1.0L with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the cultivation of microorganisms and detection of pectate-degrading enzymes.

**Polysorbate 80 Agar
(Tween 80 Agar)**

Composition per 1010.0mL:

Agar	15.0g
Peptone	10.0g
Tween 80	10.0mL
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components, except Tween 80, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 45°–50°C. Autoclave for 20 min at 15 psi pressure–121°C. Pour into sterile Petri dishes

Use: For the cultivation of a variety of microorganisms.

Pork Plasma Fibrinogen Overlay Agar

Composition per plate:

Baird-Parker agar, modified..... 15.0mL
Pork plasma fibrinogen overlay agar 8.0mL
pH 7.0 ± 0.1 at 25°C

Baird-Parker Agar, Modified:

Composition per liter:

Agar 17.0g
Glycine 12.0g
Sodium pyruvate 10.0g
Pancreatic digest of casein 10.0g
Beef extract 5.0g
LiCl 5.0g
Yeast extract 1.0g
Chapman tellurite solution 1.0mL
pH 7.0 ± 0.2 at 25°C

Chapman Tellurite Solution:

Composition per 100.0mL:

K₂TeO₃ 1.0g

Preparation of Chapman Tellurite Solution:

Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Baird-Parker Agar, Modified:

Add components, except Chapman tellurite solution, to distilled/deionized water and bring volume to 999.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45–50°C. Aseptically add 1.0mL of sterile Chapman tellurite solution. Mix thoroughly but gently.

Pork Plasma Fibrinogen Overlay Agar:

Composition per 100.5mL:

Agar solution 50.0mL
Bovine fibrinogen solution 47.5mL
Pork plasma 2.5mL
Trypsin inhibitor solution 0.5mL

Agar Solution:

Composition per 50.0mL:

Agar 0.7g

Preparation of Agar Solution: Add agar to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Bovine Fibrinogen Solution:

Composition per 50.0mL:

Bovine fibrinogen, fraction I 0.4g
Sodium phosphate buffer
(0.05M solution, pH 7.0) 50.0mL

Preparation of Bovine Fibrinogen Solution:

Grind bovine fibrinogen in a mortar to a fine powder. Add bovine fibrinogen to 50.0mL of sodium phosphate buffer. Mix thoroughly on a magnetic stirrer for 30 min. Filter through Whatman #1 filter paper. Filter sterilize.

Pork Plasma:

Composition per 10.0mL:

Pork plasma-EDTA 10.0mL

Preparation of Pork Plasma: Filter sterilize fresh or rehydrated commercial pork plasma-EDTA.

Trypsin Inhibitor Solution:

Composition per 5.0mL:

Trypsin inhibitor 0.015g
Sodium phosphate buffer
(0.05M solution, pH 7.0) 5.0mL

Preparation of Trypsin Inhibitor Solution:

Add trypsin inhibitor to 5.0mL of sodium phosphate buffer. Mix thoroughly. Filter sterilize.

Preparation of Pork Plasma Fibrinogen Overlay Agar:

Aseptically combine 50.0mL of cooled, sterile agar solution, 47.5mL of sterile bovine fibrinogen solution, 2.5mL of sterile pork plasma, and 0.5mL of trypsin inhibitor solution. Mix thoroughly. Maintain at 45°–50°C but use within 1 hr.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Pour cooled, sterile Baird-Parker agar, modified, into sterile Petri dishes in 15.0mL volumes. Allow agar to solidify. Overlay each plate with 8.0mL of sterile pork plasma fibrinogen overlay agar.

Use: For the cultivation of *Staphylococcus aureus* from foods.

Potassium Cyanide Broth (BAM M126)

Composition per liter:

Na₂HPO₄ 5.64g
NaCl 5.0g
Proteose peptone No. 3 3.0g
KH₂PO₄ 0.225g
KCN solution 15.0mL
pH 7.6 ± 0.2 at 25°C

KCN Solution:**Composition** per 100.0mL:

KCN 0.5g

Preparation of KCN Solution: Add KCN to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Caution: Cyanide is toxic. Do not pipet by mouth. Use protective equipment/gloves when handling.

Preparation of Medium: Add components, except KCN solution, to distilled/deionized water and bring volume to 985.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 15.0mL of KCN solution. Mix thoroughly. Distribute into sterile screw-capped tubes or flasks in 1.0–1.5mL volumes. Close caps tightly. Store at 5–8°C no longer than 2 weeks before use.

Use: For the cultivation and differentiation of urease-negative, Gram-negative enteric bacteria. *Salmonella* species and *Shigella* species are nonmotile in this medium. *Proteus* species are motile in this medium.

Potassium Tellurite Agar**Composition** per liter:

Beef heart, solids from infusion 500.0g

Agar 15.0g

Tryptose 10.0g

NaCl 5.0g

K₂TeO₃ solution 20.0mL

Blood, defibrinated 50.0mL

pH 6.0 ± 0.2 at 25°C

K₂TeO₃ Solution:**Composition** per 20.0mL:K₂TeO₃ 0.5g

Preparation of K₂TeO₃ Solution: Add K₂TeO₃ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components, except K₂TeO₃ solution, to distilled/deionized water and bring volume to 930.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile K₂TeO₃ solution and 50.0mL of blood. Rabbit or sheep blood may be used. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of *Enterococcus faecalis*. *Enterococcus faecalis* appears as black colonies.

Potato Agar with Cereal Culms**Composition** per liter:

Potatoes 200.0g

Agar 20.0g

Cereal culms variable

Preparation of Medium: Peel and dice potatoes. Add potatoes to 1.0L of tap water. Gently heat and bring to boiling. Boil potatoes until thoroughly cooked. Filter through cheesecloth. Bring volume of filtrate to 1.0L with tap water. Add 20.0g of agar. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Cut cereal culms into 1.0cm pieces. Place 6 to 9 cereal culm pieces in tubes with 3.0mL of distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Melt agar tubes by gently heating. Cool to 50°C. Aseptically add 2 to 3 sterile cereal culm pieces to each tube. Allow to solidify in a slanted position.

Use: For the cultivation of fungi.

**Potato Dextrose Adenine Agar
(ATCC Medium 2204)****Composition** per liter:

Glucose 20.0g

Agar 15.0g

Potatoes, infusion from 500.0mL

Adenine solution 50.0mL

pH 5.6 ± 0.2 at 25°C

Potato Infusion:**Composition** per 500.0mL:

Potatoes 300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Adenine Solution:**Composition** per 50.0mL:

Adenine 0.1g

Preparation of Adenine Solution: Add adenine to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except adenine solution to distilled/deionized water and bring volume to 950.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile adenine solution. Mix thoroughly. Pour into sterile Petri dishes or distribute to sterile tubes.

Use: For the cultivation of yeasts and molds.

Potato Dextrose Agar

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potato, infusion from	4.0g
Tartaric acid solution	14.0mL
pH 5.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Tartaric Acid Solution:

Composition per 50.0mL:

Tartaric acid	5.0g

Preparation of Tartaric Acid Solution: Add tartaric acid to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 986.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 14.0mL of sterile tartaric acid solution. Mix thoroughly. If medium is to be used for the enumeration of yeasts and molds in butter, adjust pH to 3.5. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and enumeration of yeasts and molds. For the enumeration of yeasts and molds in butter by the plate count method.

Potato Dextrose Agar (BAM M127)

Composition per liter:

Agar	20.0g
Glucose	20.0g
Potato infusion	1.0L
pH 5.6 ± 0.2 at 25°C	

Potato Infusion:

Composition per 1.0L:

Potatoes, unpeeled and sliced	200.0g

Preparation of Potato Infusion: Add unpeeled potato slices to 1.0L of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate. Bring volume to 1.0L with distilled/deionized water.

Preparation of Medium: Add agar and glucose to 1.0L potato infusion. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of yeasts and filamentous fungi (molds) from foods.

Potato Dextrose Agar (PDA)

Composition per liter:

Potatoes.....	300.0g
Glucose	20.0g
Agar	15.0g

Preparation of Medium: Dice potatoes and place in 500.0mL of boiling water for 30 min. Strain through cheesecloth. Adjust volume to 1.0L with distilled/deionized water. Mix thoroughly. Add agar. Gently heat and bring to boiling. Add 20.0g of glucose. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of numerous fungi.

Potato Dextrose Agar (PDA Agar) (ATCC Medium 336)

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potatoes, infusion from.....	500.0mL
pH 5.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Potato Infusion:

Composition per 500.0mL:

Potatoes.....	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds from dairy products and other foods. Also used to induce sporulation in many fungi.

Potato Dextrose Agar, pH 5.0
(PDA, pH5.5)
(ATCC Medium 368)

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potatoes, infusion from	500.0mL
pH 5.0 ± 0.2 at 25°C	

Potato Infusion:**Composition per 500.0mL:**

Potatoes	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.0. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds from dairy products and other foods.

Potato Dextrose Agar, 1/3 Strength
(PDA, 1/3 Strength)
(ATCC Medium 2369)

Composition per liter:

Glucose	6.67g
Agar	15.0g
Potatoes, infusion from	333.0mL
pH 5.6 ± 0.2 at 25°C	

Potato Infusion:**Composition per 1.0L:**

Potatoes	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Bring volume to 1.0L. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds.

Potato Dextrose Agar, Quarter Strength
(PDA, Quarter Strength)
(ATCC Medium 2218)

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potatoes, infusion from	125.0mL
pH 5.6 ± 0.2 at 25°C	

Potato Infusion:**Composition per 500.0mL:**

Potatoes	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 60 min. Filter through cheesecloth. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds from dairy products and other foods.

Potato Dextrose Agar and Yeast Medium
(PDA and Yeast Medium)
(ATCC Medium 104)

Composition per liter:

Glucose	20.0g
Agar	15.0g
KH ₂ PO ₄	2.0g
Yeast extract	1.5g
MgSO ₄ ·7H ₂ O	0.5g
Potato infusion	500.0mL

Potato Infusion:**Composition per 500.0mL:**

Potatoes	300.0g

Preparation of Potatoes, Infusion From: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Pseudomonas fluorescens*, *Streptovorticillium baldacii*, and *Xanthomonas campestris*.

Potato Dextrose Agar with Antibiotics

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potato, infusion from	4.0g
Antibiotic solution	20.0mL

Antibiotic Solution:

Composition per 100.0mL:

Chlortetracycline-HCl	0.5g
Chloramphenicol	0.5g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 20.0mL of sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of fungi from foods.

Potato Dextrose Agar with Gentamycin (ATCC Medium 2286)

Composition per liter:

Glucose	20.0g
Agar	15.0g
Potatoes, infusion from	500.0mL
pH 5.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Potato Infusion:

Composition per 500.0mL:

Potatoes	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Gentamycin Solution:

Composition per 10.0mL:

Gentamycin	100mg

Preparation of Gentamycin Solution: Add gentamycin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except gentamycin to distilled/deionized water and bring volume to 990.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add

10.0mL of sterile gentamycin solution. Mix thoroughly. Pour into sterile Petri dishes or sterile tubes.

Use: For the cultivation of yeasts and molds from dairy products and other foods.

Potato Dextrose Agar with 2% Glucose and 60% Sucrose

Composition per liter:

Sucrose	600.0g
Glucose	20.0g
Agar	15.0g
Potato, solids from infusion	4.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Saccharomyces rouxii* from chocolate syrup.

Potato Dextrose Agar with Thiamine (ATCC Medium 2391)

Composition per liter:

Glucose	20.0g
Agar	15.0g
Thiamine-HCl	2.0mg
Potatoes, infusion from	500.0mL
pH 5.6 ± 0.2 at 25°C	

Potato Infusion:

Composition per 500.0mL:

Potatoes	300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 60 min. Filter through cheesecloth. Bring volume to 1.0L with distilled/deionized water. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds.

Potato Dextrose Broth

Composition per liter:

Potatoes, infusion from	200.0g
Glucose	20.0g
pH 5.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Potatoes, Infusion From:

Composition per 500.0mL:

Potatoes 300.0g

Preparation of Potatoes, Infusion From: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of yeasts and molds.

Potato Dextrose Broth with Yeast Extract

Composition per liter:

Potato, peeled and cut 200.0g

Glucose 10.0g

Yeast extract 3.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Peel and cut potatoes. Add potatoes to 500.0mL of water. Boil potatoes for 20 min. Filter through cheesecloth. Add glucose and yeast extract to filtrate. Bring volume to 1.0L with distilled/deionized water. Mix thoroughly. Adjust pH to 7.2. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of fungi.

Potato Dextrose L-Isoleucine Agar (ATCC Medium 2205)

Composition per liter:

Glucose 20.0g

Agar 15.0g

Potatoes, infusion from 500.0mL

L-Isoleucine solution 50.0mL

pH 5.6 ± 0.2 at 25°C

Potato Infusion:

Composition per 500.0mL:

Potatoes 300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

L-Isoleucine Solution:

Composition per 50.0mL:

L-Isoleucine 0.13g

Preparation of L-Isoleucine Solution: Add L-isoleucine to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-isoleucine solution to distilled/deionized water and bring volume to 950.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile L-isoleucine solution. Mix thoroughly. Pour into sterile Petri dishes or distribute to sterile tubes.

Use: For the cultivation of yeasts and molds.

Potato Dextrose Salt Agar (BAM M127)

Composition per liter:

NaCl 75.0g

Agar 20.0g

Glucose 20.0g

Potato infusion 1.0L

pH 5.6 ± 0.2 at 25°C

Potato Infusion:

Composition per 1.0L:

Potatoes, unpeeled and sliced 200.0g

Preparation of Potato Infusion: Add unpeeled potato slices to 1.0L of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate. Bring volume to 1.0L with distilled/deionized water.

Preparation of Medium: Add agar and glucose to 1.0L potato infusion. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of halophilic yeasts and halophilic filamentous fungi (molds) from foods.

Potato Dextrose Yeast Agar (PDY Agar)

Composition per liter:

Glucose 20.0g

Agar 15.0g

Yeast extract 5.0g

Potato infusion 500.0mL

pH 5.6 ± 0.2 at 25°C

Potato Infusion:**Composition** per 500.0mL:

Potatoes 300.0g

Preparation of Potato Infusion: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth. Reserve filtrate.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* species and fungi; also used to induce sporulation in many fungi.

Potato Extract Agar**Composition** per liter:

Agar 15.0g
 Peptone 5.0g
 NaCl 5.0g
 Yeast extract 2.0g
 Beef extract powder 1.0g
 Potato extract 20.0mL

pH 7.4 ± 0.2 at 25°C

Potato Extract:**Composition** per liter:

Potatoes 300.0g

Preparation of Potato Extract: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth.

Use: For the cultivation of a wide variety of yeasts and molds.

Potato Flakes Agar**Composition** per liter:

Potato flakes 20.0g
 Agar 15.0g
 Glucose 10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and induction of sporulation in all fungi.

Potato Malt Agar**Composition** per liter:

Potatoes, infusion from 200.0g
 Sucrose 60.0g
 Agar 20.0g
 Malt extract 20.0g
 Peptone 1.0g

pH 7.4 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Potatoes, Infusion From:**Composition** per 500.0mL:

Potatoes 300.0g

Preparation of Potatoes, Infusion From: Peel and dice potatoes. Add 500.0mL of distilled/deionized water. Gently heat and bring to boiling. Continue boiling for 30 min. Filter through cheesecloth.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fungi and other aciduric microorganisms.

Powell and Errington's Medium**Composition** per 1060.0mL:

Solution 1 50.0mL
 Solution 3 50.0mL
 Solution 4 10.0mL
 Solution 2 5.0mL

pH 7.0 ± 0.2 at 25°C

Solution 1:**Composition** per liter:

(NH₄)₂HPO₄ 238.0g
 K₂SO₄ 70.0g
 NaH₂PO₄·2H₂O 31.0g

Preparation of Solution 1: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Solution 2:**Composition** per liter:

MgO 10.0g
 FeCl₃·6H₂O 5.4g
 CaCO₃ 2.0g
 ZnSO₄·7H₂O 1.44g
 MnSO₄·4H₂O 1.11g
 Na₂MoO₄·2H₂O 0.49g
 CoSO₄·7H₂O 0.28g
 CuSO₄·5H₂O 0.25g

H ₃ BO ₄	0.062g
HCl, concentrated	50.0mL

Preparation of Solution 2: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Solution 3:

Composition per 50.0mL:

Citric acid	4.2g
Glucose	3.6g
L-Glutamic acid	2.94g
Succinic acid	1.18g

Preparation of Solution 3: Add components to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Solution 4:

Composition per 10.0mL:

Na ₂ S ₂ O ₃ ·5H ₂ O	1.24g

Preparation of Solution 4: Add Na₂S₂O₃·5H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add 50.0mL of solution 1 and 5.0mL of solution 2. Mix thoroughly. Bring volume to 1.0L with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Adjust pH to 7.0 with sterile NaOH. Aseptically add 50.0mL of solution 3 and 10.0mL of sterile solution 4. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of a variety of heterotrophic microorganisms.

Preenrichment Medium (PEM)

Composition per liter:

Yeast extract	20.0g
Special peptone, Oxoid	10.0g
Na ₂ HPO ₄	7.1g
NaCl	1.0g
KCl	1.0g
MgSO ₄ ·7H ₂ O solution	10.0mL
CaCl ₂ ·2H ₂ O solution	10.0mL
pH 8.3 ± 0.2 at 25°C	

MgSO₄·7H₂O Solution:

Composition per 10.0mL:

MgSO ₄ ·7H ₂ O	0.01g

Preparation of MgSO₄·7H₂O Solution: Add MgSO₄·7H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

CaCl₂·2H₂O Solution:

CaCl ₂ ·2H ₂ O	0.01g

Preparation of CaCl₂·2H₂O Solution: Add the CaCl₂·2H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except MgSO₄·7H₂O solution and CaCl₂·2H₂O solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 8.3. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile MgSO₄·7H₂O solution and CaCl₂·2H₂O solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the isolation and enrichment of *Yersinia enterocolitica* from foods.

Preston Enrichment Broth

Composition per liter:

Beef extract	10.0g
Peptone	10.0g
NaCl	5.0g
Horse blood, lysed	50.0mL
Antibiotic solution	10.0mL
pH 7.5 ± 0.2 at 25°C	

Antibiotic Solution:

Composition per 10.0mL:

Cycloheximide	0.1g
Rifampicin	0.01g
Trimethoprim lactate	0.01g
Polymyxin B	5000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse blood and antibiotic solution, to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile horse blood and antibiotic solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and enrichment of *Campylobacter* species from foods.

Presumptive Media

Composition per plate:

Quadrant I	5.0mL
Quadrant II	5.0mL
Quadrant III	5.0mL
Quadrant IV	5.0mL

Quadrant I:**Composition per 5.0mL:**

Lombard-Dowell agar 5.0mL

Quadrant II:**Composition per 5.0mL:**

Lombard-Dowell bile agar 5.0mL

Quadrant III:**Composition per 5.0mL:**

Lombard-Dowell egg yolk agar 5.0mL

Quadrant IV:**Composition per 5.0mL:**

Lombard-Dowell esculin agar 5.0mL

Use: For the differentiation and presumptive identification of anaerobic bacteria. The Presumpto media is a four-sectored plate. Each plate contains a different medium. See Lombard-Dowell agars for additional information.

Pril Xylose Ampicillin Agar (PXA Agar)

Composition per liter:

Agar 15.0g

Xylose 10.0g

Pancreatic digest of gelatin 5.0g

Beef extract 3.0g

Pril 0.2g

Ampicillin 0.03g

Phenol Red 0.025g

pH 6.8 ± 0.2 at 25°C

Note: Pril is a quaternary ammonium detergent composed of a mixture of primary alkyl sulfate, alkyl-benzyl sulfonate and salts. It is available from Böhme Fettchemie GmbH, Düsseldorf, Germany.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation and cultivation of *Aeromonas hydrophila*.

Propionibacterium Agar

Composition per liter:

Agar 15.0g

Casein peptone, tryptic digest 10.0g

Sodium lactate 10.0g

Yeast extract 5.0g

pH 7.0–7.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0–7.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Propionibacterium acidipropionici* and *Propionibacterium thoenii*.

Propionibacterium Medium

Composition per liter:

Yeast extract 10.0g

Na₂HPO₄·2H₂O 3.0gKH₂PO₄ 1.0g

Lactate solution 40.0mL

pH 7.0 ± 0.2 at 25°C

Lactate Solution:**Composition per 100.0mL:**

Sodium lactate 70.0g

Preparation of Lactate Solution: Add 70.0g of sodium lactate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except lactate solution, to distilled/deionized water and bring volume to 960.0mL. Mix thoroughly. Add 40.0mL of lactate solution. Mix thoroughly. Adjust pH to 7.0. Distribute into screw-capped tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Propionibacterium acidipropionici*, *Propionibacterium freudenreichii*, *Propionibacterium jensenii*, and *Propionibacterium thoenii*.

Proskauer-Beck Medium for *Mycobacterium*

Composition per liter:

Asparagine 5.0g

KH₂PO₄ 5.0g

Magnesium citrate 2.5g

MgSO₄·7H₂O 0.6g

Glycerol 20.0mL

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components, one at a time, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Make sure one salt is totally dissolved before the next one is added. Adjust pH to 7.8 with 40% NaOH. Autoclave for 15 min at 15 psi pressure–121°C. The pH of the medium after autoclaving should be 7.4. Filter through Whatman #1 filter paper to remove any precipitate. Distribute

into tubes or flasks. Autoclave again for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Mycobacterium tuberculosis*.

Proteose Agar

Composition per liter:

Agar	15.0g
Proteose peptone No. 3	15.0g
Yeast extract	7.5g
Casamino acids	5.0g
K ₂ HPO ₄	5.0g
(NH ₄) ₂ SO ₄	1.5g
Starch, soluble	1.0g

pH 9.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the cultivation of *Vibrio* species from foods.

Proteose No. 3 Agar (ATCC Medium 50)

Composition per 1010.0mL:

Proteose peptone No. 3	20.0g
Agar	15.0g
Na ₂ HPO ₄	5.0g
NaCl	5.0g
Glucose	0.5g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli* and other bacteria.

Proteose Yeast Extract Medium

Composition per liter:

Proteose peptone	20.0g
Glucose	10.0g
Yeast extract	5.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of bacteria.

***Pseudomonas aeruginosa* Agar**

(PA Agar)

(m–PA Agar)

(m–*Pseudomonas aeruginosa* Agar)

Composition per liter:

Agar	15.0g
Na ₂ S ₂ O ₃	6.8g
L-Lysine·HCl	5.0g
NaCl	5.0g
Xylose	2.5g
Yeast extract	2.0g
Lactose	1.25g
Sucrose	1.25g
Ferric ammonium citrate	0.8g
Sulfapyridine	0.176g
Cycloheximide	0.15g
Phenol Red	0.08g
Nalidixic acid	0.037g
Kanamycin	8.5mg

pH 7.1 ± 0.2 at 25°C

Preparation of Medium: Add components—except sulfapyridine, cycloheximide, nalidixic acid, and kanamycin—to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.5. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 55°–60°C. Readjust pH to 7.1. Aseptically add the sulfapyridine, cycloheximide, nalidixic acid, and kanamycin. Mix thoroughly. Pour into 50mm × 12mm Petri dishes in 3.0mL volumes.

Use: For the cultivation and estimation of numbers of *Pseudomonas aeruginosa* by the membrane filter method.

***Pseudomonas* Agar F**

(BAM M128)

Composition per liter:

Proteose peptone No. 3	20.0g
Agar	15.0g
Glycerol	10.0g
Pancreatic digest of casein	10.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	0.73g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and observation of fluorescein production in *Pseudomonas* species.

***Pseudomonas* Agar F**

Composition per liter:

Agar	15.0g
Glycerol	10.0g
Proteose peptone No. 3	10.0g
Pancreatic digest of casein	10.0g
K ₂ HPO ₄	1.5g
MgSO ₄ ·7H ₂ O	1.5g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and differentiation of *Pseudomonas aeruginosa* on the basis of pigment production.

***Pseudomonas* Agar P (BAM M129)**

Composition per liter:

Proteose peptone No. 3	20.0g
Agar	15.0g
Glycerol	10.0g
K ₂ HPO ₄	10.0g
MgCl ₂ ·6H ₂ O	1.4g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and differentiation of *Pseudomonas aeruginosa* on the basis of pigment production.

***Pseudomonas* Basal Mineral Medium**

Composition per liter:

K ₂ HPO ₄	12.5g
KH ₂ PO ₄	3.8g
(NH ₄) ₂ SO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.1g
Carbon source (0.8M solution)	100.0mL
Trace element solution	5.0mL

pH 7.2 ± 0.2 at 25°C

Trace Element Solution:

Composition per liter:

H ₃ BO ₃	0.232g
ZnSO ₄ ·7H ₂ O	0.174g
FeSO ₄ (NH ₄) ₂ SO ₄ ·6H ₂ O	0.116g
CoSO ₄ ·7H ₂ O	0.096g
(NH ₄) ₆ Mo ₇ O ₂₄ ·4H ₂ O	0.022g
CuSO ₄ ·5H ₂ O	8.0mg
MnSO ₄ ·4H ₂ O	8.0mg

Preparation of Trace Element Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Carbon Source:

Composition per 100.0mL:

Glucose	14.4g

Preparation of Carbon Source: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Other carbon sources may replace glucose. Prepare 0.8M carbon source solution.

Preparation of Medium: Add components, except carbon source, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile carbon source. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and differentiation of *Pseudomonas* species based on their ability to grow on different carbon sources.

***Pseudomonas* CFC Agar**

Composition per liter:

Pancreatic digest of gelatin	16.0g
Agar	11.0g
Pancreatic digest of casein	10.0g
K ₂ SO ₄	10.0g
MgCl ₂ ·6H ₂ O	1.4g

CFC selective supplement	10.0mL
Glycerol	10.0mL
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

CFC Selective Supplement:

Composition per 10.0mL:

Cephaloridine.....	0.05g
Fucidin	0.01g
Cetrimide	0.01g

Preparation of CFC Selective Supplement:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except CFC selective supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile CFC selective supplement. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Pseudomonas* species.

***Pseudomonas* CN Agar**

Composition per liter:

Pancreatic digest of gelatin.....	16.0g
Agar	11.0g
Pancreatic digest of casein.....	10.0g
K ₂ SO ₄	10.0g
MgCl ₂ ·6H ₂ O	1.4g
CN selective supplement	10.0mL
Glycerol	10.0mL

CN Selective Supplement:

Cetrimide	0.1g
Sodium nalidixate	7.5mg
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except CN selective supplement, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile CN selective supplement. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Pseudomonas* species.

***Pseudomonas* Isolation Agar**

Composition per liter:

Peptone	20.0g
Agar	13.6g
K ₂ SO ₄	10.0g
MgCl ₂ ·6H ₂ O	1.4g
Irgasan (triclosan)	0.025g
Glycerol	20.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Pseudomonas* species.

***Pseudomonas* Medium (ATCC Medium 59)**

Composition per liter:

K ₂ HPO ₄	1.15g
NH ₄ NO ₃	1.0g
Yeast extract.....	1.0g
KH ₂ PO ₄	0.625g
MgSO ₄ ·7H ₂ O	0.02g
Pyrrolidine	4.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add pyrrolidine to approximately 500.0mL of distilled/deionized water. Mix thoroughly. Adjust pH to 7.0. Add remaining components. Bring volume to 1.0L with distilled/deionized water. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Pseudomonas fluorescens*.

***Pseudomonas* Medium (ATCC Medium 609)**

Composition per liter:

Agar	15.0g
K ₂ HPO ₄	8.71g
Nitrilotriacetic acid	1.91g
Na ₂ SO ₄	0.57g
MgSO ₄	0.25g
FeSO ₄	0.5mg
Ca(NO ₃) ₂	0.5mg
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add nitrilotriacetic acid to approximately 500.0mL of distilled/deionized water. Mix thoroughly. Adjust pH to 6.5. Add remaining components. Bring volume to 1.0L with distilled/deionized water. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Pseudomonas* species.

***Pseudomonas* Medium (ATCC Medium 775)**

Composition per liter:

NH ₄ Cl	5.0g
K ₂ HPO ₄	1.5g
L-Tryptophan	1.0g
KH ₂ PO ₄	0.5g
Yeast extract	0.5g
MgSO ₄	0.2g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Comamonas acidovorans*.

***Pseudomonas* Medium A**

Composition per liter:

Peptone	20.0g
Agar	15.0g
Glycerol	10.0g
K ₂ SO ₄	10.0g
MgCl ₂	1.4g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–115°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and production of pyocyanin by *Pseudomonas* species.

***Pseudomonas* Medium B**

Composition per liter:

Peptone	20.0g
Agar	15.0g
Glycerol	10.0g

MgSO ₄ ·7H ₂ O	1.5g
KH ₂ PO ₄ solution	100.0mL
pH 7.2 ± 0.2 at 25°C	

K₂HPO₄ Solution:

Composition per 100.0mL:

KH ₂ PO ₄	1.5g

Preparation of K₂HPO₄ Solution: Add K₂HPO₄ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components, except K₂HPO₄ solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile K₂HPO₄ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and observation of fluorescein production by *Pseudomonas* species.

***Pseudomonas saccharophila* Medium**

Composition per 1015.0mL:

Agar	20.0g
Na ₂ HPO ₄	4.8g
KH ₂ PO ₄	4.4g
NH ₄ Cl	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Solution A	5.0mL
Solution B	10.0mL

Solution A:

Composition per 100.0mL:

Ferric ammonium citrate	1.0g
CaCl ₂	0.1g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Solution B:

Composition per 100.0mL:

Sucrose	10.0g

Preparation of Solution B: Add sucrose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except solution A and solution B, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile solution A and sterile solution B. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Pseudomonas saccharophila* and other *Pseudomonas* species.

Purple Agar

Composition per liter:

Agar	15.0g
Proteose peptone No. 3	10.0g
NaCl	5.0g
Beef extract	1.0g
Bromcresol Purple	0.02g
Carbohydrate solution	20.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. For expensive carbohydrates, 5.0g may be used instead of 10.0g. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 9.8mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube. Mix thoroughly. Allow tubes to cool in a slanted position.

Use: For the preparation of carbohydrate media used in fermentation studies for the identification of bacteria, especially members of the *Enterobacteriaceae*. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Broth (Purple Carbohydrate Broth) (BAM M130)

Composition per liter:

Proteose peptone No. 3	10.0g
NaCl	5.0g
Beef extract	1.0g
Bromcresol Purple	0.02g
Carbohydrate solution	20.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. For expensive carbohydrates, 5.0g may be used instead of 10.0g. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 9.8mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube. Mix thoroughly.

Use: For the preparation of carbohydrate media used in fermentation studies for the identification of bacteria, especially members of the *Enterobacteriaceae*. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Broth

Composition per liter:

Pancreatic digest of gelatin	10.0g
NaCl	5.0g
Bromcresol Purple	0.02g
Carbohydrate solution	20.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. For expensive carbohydrates, 5.0g may be used instead of 10.0g. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 9.8mL of medium. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube.

Use: For the preparation of liquid fermentation media. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Broth with NaCl
(Purple Carbohydrate Broth with NaCl)
(BAM M130)

Composition per liter:

Proteose peptone No. 3	10.0g
NaCl.....	5.0g
Beef extract.....	1.0g
Bromcresol Purple	0.02g
Carbohydrate solution.....	20.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Carbohydrate Solution:

Composition per 20.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 20.0mL. For expensive carbohydrates, 5.0g may be used instead of 10.0g. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 9.8mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 0.2mL of sterile carbohydrate solution to each tube. Mix thoroughly.

Use: For the preparation of carbohydrate media used in fermentation studies for the identification of bacteria, especially members of the *Enterobacteriaceae*. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Carbohydrate Fermentation
Broth Base
(BAM M130a)

Composition per liter:

Pancreatic digest of gelatin.....	10.0g
NaCl.....	5.0g
Bromcresol Purple	0.02g
Carbohydrate solution.....	100.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostics.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	5.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 9.0mL of medium. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 1.0mL of sterile carbohydrate solution to each tube.

Use: For the preparation of liquid fermentation media. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Carbohydrate Fermentation
Broth Base with Esculin
(BAM M130a)

Composition per liter:

Pancreatic digest of gelatin.....	10.0g
NaCl.....	5.0g
Esculin	5.0g
Bromcresol Purple	0.02g
Carbohydrate solution.....	10.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium without esculin is available as a premixed powder from BD Diagnostics.

Carbohydrate Solution:

Composition per 100.0mL:

Carbohydrate.....	5.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution and esculin, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.4 if necessary. Distribute into tubes containing an inverted Durham tube. Fill each tube with 9.0mL of medium. Add 0.05g esculin to each tube. Autoclave for 15 min at 10 psi pressure–115°C. Aseptically add 1.0mL of sterile carbohydrate solution to each tube.

Use: For the preparation of liquid fermentation media, e.g., for *Listeria* spp. and *Enterococcus* spp. Bacteria that can ferment the carbohydrate turn the medium yellow.

Purple Lactose Agar**Composition** per liter:

Agar	10.0g
Lactose	10.0g
Peptone.....	5.0g
Beef extract.....	3.0g
Bromcresol Purple	0.025g

pH 6.8 ± 0.1 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the detection and differentiation of halophilic *Vibrio* spp. Bacteria that can ferment lactose turn the medium yellow.

Purple Serum Agar Base**Composition** per liter:

Agar	20.0g
Lactose	20.0g
Peptone.....	20.0g
NaCl.....	5.0g
Bromcresol Purple	0.03g
Phenol Red.....	0.024g

pH 7.6 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and differentiation of Gram-negative bacteria isolated from the urinary tract. Bacteria that can ferment lactose turn the medium yellow.

**PY Medium with Glucose
(Peptone Yeast Extract
Medium with Glucose)
(PYG Medium)**

Composition per liter:

Glucose	10.0g
Yeast extract.....	10.0g
Peptone.....	5.0g
Pancreatic digest of casein.....	5.0g

L-cysteine·HCl·H ₂ O	0.5g
Salts solution.....	40.0mL
Hemin solution.....	10.0mL
Resazurin solution	4.0mL
Vitamin K ₁ solution	0.2mL

pH 6.9 ± 0.2 at 25°C

Salts Solution:**Composition** per liter:

NaHCO ₃	10.0g
NaCl.....	2.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
CaCl ₂ , anhydrous	0.2g
MgSO ₄	0.2g

Preparation of Salts Solution: Add CaCl₂ and MgSO₄ to 300.0mL of distilled/deionized water. Mix thoroughly until dissolved. Bring volume to 800.0mL with distilled/deionized water. Add remaining components while stirring. Bring volume to 1.0L. Mix thoroughly. Store at 4°C.

Hemin Solution:**Composition** per 100.0mL:

Hemin	0.05g
NaOH (1N solution).....	1.0mL

Preparation of Hemin Solution: Add hemin to NaOH solution. Mix thoroughly. Adjust volume to 100.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Resazurin Solution:**Composition** per 44.0mL:

Resazurin	0.044g

Preparation of Resazurin Solution: Add resazurin to distilled/deionized water and bring volume to 44.0mL. Mix thoroughly.

Vitamin K₁ Solution:**Composition** per 30.0mL:

Vitamin K ₁	0.15g
Ethanol (95% solution).....	30.0mL

Preparation of Vitamin K₁ Solution: Add vitamin K₁ to ethanol. Mix thoroughly. Store in brown bottle and keep under refrigeration. Discard after 1 month.

Preparation of Medium: Add components—except L-cysteine·HCl·H₂O, hemin solution, and vitamin K₁ solution—to distilled/deionized water and bring volume to 989.8mL. Mix thoroughly. Gently heat and bring to boiling under 80% N₂ + 10% CO₂ + 10% H₂. Continue boiling until resazurin turns colorless, indicating reduction. Cool to 45°–50°C. Add the L-cysteine·HCl·H₂O, hemin solution, and vitamin

K₁ solution. Adjust pH to 6.9 if necessary. Anaerobically distribute into tubes under 80% N₂ + 10% CO₂ + 10% H₂. Cap the tubes with rubber stoppers. Place tubes in a press. Autoclave for 15 min at 15 psi pressure–121°C with fast exhaust.

Use: For the cultivation and maintenance of *Clostridium* species.

PYG Broth

(Peptone Yeast Extract Glucose Broth)

Composition per liter:

Peptone.....	20.0g
D-Glucose.....	10.0g
Yeast extract.....	10.0g
L-Cysteine·HCl·H ₂ O.....	0.5g
VPI salt solution.....	40.0mL
Resazurin solution.....	4.0mL

pH 7.2 ± 0.2 at 25°C

Resazurin Solution:

Composition per 44.0mL:

Resazurin	0.044g

Preparation of Resazurin Solution: Add resazurin to distilled/deionized water and bring volume to 44.0mL. Mix thoroughly.

VPI Salt Solution:

Composition per 40.0mL:

CaCl ₂	0.2g
MgSO ₄	0.2g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g

Preparation of VPI Salt Solution: Add CaCl₂ and MgSO₄ to 300.0mL of distilled/deionized water. Mix thoroughly until dissolved. Bring volume to 800.0mL with distilled/deionized water. Add remaining components while stirring. Bring volume to 1.0L. Mix thoroughly. Store at 4°C.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 7.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C under 100% N₂.

Use: For the cultivation of a wide variety of anaerobic bacteria.

Pyrazinamidase Agar (BAM M131)

Composition per liter:

Pancreatic digest of casein.....	11.25g
Agar	11.25g
Papaic digest of soybean meal.....	3.75g

NaCl.....	3.75g
Yeast extract.....	3.0g
Pyrazine-carboxamide	1.0g
Tris maleate, 0.2M, pH6.0	1.0L

pH 6.0 ± 0.2 at 25°C

Preparation of Medium: Add components to 0.2M tris maleate and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. For slants allow tubes to cool in an inclined position.

Use: For the cultivation of *Yersinia* spp.

Pyruvate Utilization Medium

Composition per liter:

Sodium pyruvate.....	10.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	5.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Bromthymol Blue	0.1g

pH 7.1–7.4 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.1–7.4. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of bacteria that can metabolize pyruvate. Bacteria that can utilize pyruvate turn the medium yellow.

R2 Broth

Composition per liter:

Peptones.....	1.0g
Yeast extract.....	0.5g
Glucose	0.5g
Starch, soluble.....	0.5g
KH ₂ PO ₄	0.3g
Sodium pyruvate.....	0.3g
MgSO ₄ ·7H ₂ O	0.024g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus* spp. and *Deinococcus* spp.

R2A Agar**Composition** per liter:

Agar	15.0g
Yeast extract.....	0.5g
Acid hydrolysate of casein.....	0.5g
Glucose	0.5g
Soluble starch.....	0.5g
K ₂ HPO ₄	0.3g
Sodium pyruvate.....	0.3g
Pancreatic digest of casein.....	0.25g
Peptic digest of animal tissue.....	0.25g
MgSO ₄ , anhydrous.....	0.024g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not over-heat. Pour into sterile Petri dishes or leave in tubes.

Use: For use in standard methods for pour plate, spread plate, and membrane filter analysis to enumerate heterotrophic bacteria from waters.

R3A Agar**Composition** per liter:

Agar	15.0g
Yeast extract.....	1.0g
Acid hydrolysate of casein.....	1.0g
Glucose	1.0g
Soluble starch.....	1.0g
K ₂ HPO ₄	0.6g
Sodium pyruvate.....	0.6g
Pancreatic digest of casein.....	0.5g
Peptic digest of animal tissue.....	0.5g
MgSO ₄ , anhydrous.....	0.048g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not over-heat. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of heterotrophic bacteria from potable waters.

Rainbow Agar O157**Composition** per liter:

Proprietary

pH 7.9–8.3 at 25°C

Source: This medium is available from Biolog.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and boil to dissolve components. Autoclave for 10 min at 15 psi pressure–121°C. Do not exceed 10 min at this temperature and pressure. For volumes less than 500.0mL, autoclave for 5 min. Cool agar to 45–50°C before pouring plates or adding selective agents. To increase the selectivity of this medium, add a sterile solution of 0.8 mg/L potassium tellurite and 10.0 mg/L novobiocin. Tellurite is highly selective for *E. coli* O157:H7 and will reduce background flora considerably. Novobiocin inhibits *Proteus* swarming and the growth of tellurite-reducing bacteria.

Use: As a selective, chromogenic culture medium to aid in the detection, isolation, and presumptive identification of verotoxin-producing strains of *Escherichia coli*, particularly serotype O157:H7. The medium contains chromogenic substrates that are specific for two *E. coli*-associated enzymes: β-galactosidase (a blue-black chromogenic substrate) and β-glucuronidase (a red chromogenic substrate). Strain O157:H7 is typically glucuronidase negative so it forms unique and distinctive black or gray colonies. Many other non-O157 toxigenic strains overproduce β-galactosidase relative to β-glucuronidase on this medium and consequently they are typically colored purple, violet, or blue. Most non-pathogenic *E. coli* strains are glucuronidase positive, and range from pink to magenta with occasional purple or blue strains. Nearly all other bacterial species are either inhibited on this medium or they grow as white or cream-colored colonies.

Rainbow Agar Salmonella**Composition** per liter:

Proprietary

pH 7.2–7.6 at 25°C

Source: This medium is available from Biolog.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Add 10.0mL 35% glycerol. Stir until components are evenly dispersed. Gently heat and boil. Autoclave for 10 min at 15 psi pressure–121°C. Do not exceed 10 min. Cool agar to 45–50°C before pouring plates

Use: As a selective, chromogenic medium to aid in the detection and isolation of H₂S-producing *Salmo-*

nella species. Black colonies are formed by even weak H₂S-producing strains.

Raka-Ray Agar

Composition per liter:

Pancreatic digest of casein	20.0g
Agar	17.0g
Maltose	10.0g
Fructose	5.0g
Glucose	5.0g
Yeast extract	5.0g
2-Phenylethanol	3.0g
Potassium aspartate	2.5g
Potassium glutamate	2.5g
Betaine-HCL	2.0g
Diammonium hydrogen citrate	2.0g
MgSO ₄ ·7H ₂ O	2.0g
KH ₂ PO ₄	2.0g
Liver concentrate	1.0g
MnSO ₄ ·H ₂ O	0.66g
N-Acetylglucosamine	0.5g
Cycloheximide	7.0mg
Sorbitan monooleate	10.0mL
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except phenylethanol, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 3.0g of 2-phenylethanol. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation of lactic acid bacteria in beer and brewing processes. The addition of phenylethanol and cycloheximide forms a selective medium for the isolation of lactic acid bacteria in beer and brewing processes.

Rambach Agar

Composition per liter:

Agar	15.0g
Polypropylene glycol	10.5g
Peptone	8.0g
NaCl	5.0g
Na-deoxycholate	1.0g
Chromogenic mix	1.5g
pH 7.4 ± 0.2 at 25°C	

Source: Rambach Agar is available from CHROM-agar Microbiology.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat in a boiling water bath

or steam bath. Shake periodically during heating to dissolve components. Heating for 40 min with shaking every 5 min should ensure complete dissolution. Do not overheat. Cool to 45–50°C. Pour into sterile Petri dishes.

Use: For the differentiation of non-typhi *Salmonella* spp. in foodstuffs. *Salmonellae* form acid with propylene glycol and non-typhi *Salmonella* spp. form red colonies. Coliform bacteria grow as blue-green or blue-violet colonies. Other *Enterobacteriaceae* and Gram negative bacteria, such as *Proteus* spp., *Pseudomonas* spp., *Shigella* spp., *Salmonella typhi*, and *Salomonella paratyphi* A grow as colorless or yellow colonies.

Rambach® Agar

Composition per liter:

NaC	15.0g
Agar	15.0g
Propylene glycol	10.5g
Peptone	8.0g
Chromogenic mix	1.5g
Sodium deoxycholate	1.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat in a boiling water-bath or in a current of steam, while shaking from time to time. The medium is totally suspended, if no visual particles stick to the glass-wall. The medium should not be heat-treated further. Complete dissolution with shaking in 5-min sequences is approximately 35–40 min. Do not autoclave. Do not overheat. Cool as fast as possible to 45°–50°C while gently shaking from time to time. Pour into sterile Petri dishes. To prevent any precipitate or clotting of the chromogenic-mix in the plates, place Petri dishes during pouring procedure on a cool (max. 25°C) surface. The plates are opaque and pink.

Use: For the detection of enteric bacteria, including coliforms and *Salmonella* spp. Sodium deoxycholate inhibits the accompanying Gram-positive flora. This medium enables *Salmonella* spp. to be differentiated unambiguously from other bacteria. *Salmonella* spp. form a characteristic red color. In order to differentiate coliforms from *Salmonellae*, the medium contains a chromogene indicating the presence of β-galactosidase splitting, a characteristic for coliforms. Coliform microorganisms grow as blue-green or blue-violet colonies. Other *Enterobacteriaceae* and Gram-negative bacteria, such as *Proteus*, *Pseudomonas*, *Shigella*, *S. typhi*, and *S. paratyphi* A grow as colorless-yellow colonies.

RAPID'E. coli 2 Agar**Composition** per liter:

Proprietary

Source: This medium is available from Biorad.

Use: For the direct enumeration of *E. coli* and coliforms in foods. Selectivity and electivity are based on the detection of glucuronidase and galactosidase activities. Hydrolysis of chromogenic substrate results in purple to pink *E. coli* colonies (gluc+/gal+) and blue-green coliform colonies (gluc-/gal+). RAPID'E. coli 2 agar is AFNOR validated according to ISO 16140 protocol to enumerate *E. coli* and coliforms on the same plate at 37°C, without any further confirmation of characteristic colonies.

RAPID'Enterococcus Agar**Composition** per liter:

Proprietary

Source: This medium is available from Biorad.

Use: A selective chromogenic culture medium for the direct enumeration, without confirmation, of enterococci in water and in food products. The cleavage of the chromogenic substrate by glucosidase activity of Enterococci leads to specific blue colonies. RAPID'Enterococcus totally inhibits growth of Gram-negative flora and that of practically all Gram-positive bacteria other than Enterococci, due to the combined action of temperature and selective media.

**RAPID'L. mono Medium
(BAM M131a)****Composition** per liter:

Peptones	30.0g
Selective supplement, proprietary.....	20.0g
Agar B, proprietary	13.0g
D-Xylose	10.0g
LiCl	9.0g
Meat extract	5.0g
Yeast extract.....	1.0g
Phenol Red	0.12g
Chromogenic substrate, proprietary.....	1.0mL
pH 7.3 ± 0.1 at 25°C	

Source: This medium is available from Biorad.

Use: A selective chromogenic culture medium for the detection and differntiation of *Listeria* spp., including *L. ivanovii* and *L. monocytogenes*.

**Rappaport Broth, Modified
(Rap Broth, Modified)****Composition** per 250.2mL:

Solution A	155.0mL
Solution C	53.0mL
Solution B	40.0mL
Solution D	1.6mL
Solution E	0.6mL

Solution A:**Composition** per liter:

Pancreatic digest of casein.....	10.0g

Preparation of Solution A: Add pancreatic digest of casein to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Solution B:**Composition** per liter:

Na ₂ HPO ₄	9.5g

Preparation of Solution B: Add Na₂HPO₄ to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Solution C:**Composition** per 100.0mL:

MgCl ₂ ·6H ₂ O	40.0g

Preparation of Solution C: Add MgCl₂·6H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C.

Solution D:**Composition** per 100.0mL:

Malachite Green.....	0.2g

Preparation of Solution D: Add Malachite Green to sterile distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Do not sterilize.

Solution E:**Composition** per 10.0mL:

Carbenicillin	0.01g

Preparation of Solution E: Add carbenicillin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Combine 155.0mL of solution A and 40.0mL of solution B. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 53.0mL of sterile solution C, 1.6mL of solution D, and 0.6mL of sterile solution E. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the isolation and cultivation of *Yersinia enterocolitica* from foods.

**Rappaport-Vassiliadis
Enrichment Broth
(RV Enrichment Broth)
(BAM M132)**

Composition per 1110.0mL:

NaCl.....	8.0g
Papaic digest of soybean meal.....	5.0g
KH ₂ PO ₄	1.6g
Magnesium chloride solution.....	100.0mL
Malachite Green solution.....	10.0mL
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Magnesium Chloride Solution:

Composition per 100.0mL:

MgCl ₂ ·6H ₂ O	40.0g

Preparation of Magnesium Chloride Solution:

Add MgCl₂·6H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Malachite Green Solution:

Composition per 10.0mL:

Malachite Green oxalate	0.04g

Preparation of Malachite Green Solution:

Add Malachite Green to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 10 psi pressure–115°C.

Use: For the isolation and cultivation of *Salmonella* species from foods.

Rappaport-Vassiliadis R10 Broth

Composition per liter:

MgCl ₂ , anhydrous.....	13.4g
NaCl.....	7.2g
Papaic digest of soybean meal.....	4.54g
KH ₂ PO ₄	1.45g
Malachite Green oxalate	0.036g
pH 5.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 10 psi pressure–116°C.

Use: For the isolation and cultivation of *Salmonella* species from foods.

**Rappaport-Vassiliadis
Soya Peptone Broth
(RVS Broth)**

Composition per liter:

MgCl ₂ , anhydrous.....	13.58g
NaCl.....	7.2g
Papaic digest of soybean meal.....	4.5g
KH ₂ PO ₄	1.26g
K ₂ HPO ₄	0.18g
Malachite Green.....	0.036g
pH 5.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 10 psi pressure–115°C.

Use: For the isolation and cultivation of *Salmonella* species from foods.

RB-1/RB-9 Medium

Composition per liter:

Glucose or starch	10.0g
Na ₂ HPO ₄ ·12H ₂ O.....	4.2g
KH ₂ PO ₄	2.64g
Yeast extract.....	2.0g
Na ₂ SO ₄	1.0g
NH ₄ Cl	0.5g
L-Cysteine·HCl	0.5g
Na ₂ S·9H ₂ O	0.5g
MgCl ₂ ·6H ₂ O	0.36g
Resazurin	1.0mg
Trace elements solution	10.0mL
Vitamin solution.....	10.0mL
pH 5.0–6.0 at 25°C	

Vitamin Solution:

Composition per liter:

Pyridoxine·HCl	10.0mg
Calcium DL-pantothenate.....	5.0mg
Lipoic acid	5.0mg
Nicotinic acid.....	5.0mg
p-Aminobenzoic acid.....	5.0mg
Riboflavin	5.0mg
Thiamine·HCl	5.0mg
Biotin	2.0mg
Folic acid	2.0mg
Vitamin B ₁₂	0.1mg

Preparation of Vitamin Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 100% N₂. Autoclave for 15 min at 15 psi pressure–121°C.

Trace Elements Solution:

Composition per liter:

MgSO ₄ ·7H ₂ O	3.0g
Nitritotriacetic acid	1.5g
NaCl	1.0g
MnSO ₄ ·2H ₂ O	0.5g
CoSO ₄ ·7H ₂ O	0.18g
ZnSO ₄ ·7H ₂ O	0.18g
CaCl ₂ ·2H ₂ O	0.1g
FeSO ₄ ·7H ₂ O	0.1g
NiCl ₂ ·6H ₂ O	0.025g
KAl(SO ₄) ₂ ·12H ₂ O	0.02g
CuSO ₄ ·5H ₂ O	0.01g
H ₃ BO ₃	0.01g
Na ₂ MoO ₄ ·2H ₂ O	0.01g
Na ₂ SeO ₃ ·5H ₂ O	0.3mg

Preparation of Trace Elements Solution: Add nitritotriacetic acid to 500.0mL of distilled/deionized water. Adjust pH to 6.5 with KOH. Add remaining components. Add distilled/deionized water to 1.0L.

Preparation of Medium: Prepare and dispense medium under 100% N₂. Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Sparge with 100% N₂. Adjust pH to 5.0–6.0. Anaerobically distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the cultivation of *Clostridium* species.

Reinforced Clostridial Medium

Composition per liter:

Tryptose	10.0g
Beef extract	10.0g
Glucose	5.0g
NaCl	5.0g
Yeast extract	3.0g
Sodium acetate	3.0g
Soluble starch	1.0g
L-Cysteine·HCl·H ₂ O	0.5g
Agar	0.5g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 10 psi pressure–115°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the nonselective cultivation and enumeration of *Clostridium* species, other anaerobes such as lactobacilli, and facultative organisms from foods.

Reinforced Clostridial Medium with Sodium Lactate

Composition per liter:

Tryptose	10.0g
Beef extract	10.0g
Glucose	5.0g
NaCl	5.0g
Yeast extract	3.0g
Sodium acetate	3.0g
Soluble starch	1.0g
L-Cysteine·HCl·H ₂ O	0.5g
Agar	0.5g
Sodium lactate (60% solution)	15.0mL

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 10 psi pressure–115°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the nonselective cultivation and enumeration of *Clostridium* species, other anaerobes such as lactobacilli, and facultative organisms from foods.

Riboflavin Assay Medium

Composition per liter:

Peptone, photolyzed	22.0g
Glucose	20.0g
Yeast supplement	2.0g
Sodium acetate	1.8g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
L-Cystine	0.2g
FeSO ₄ ·7H ₂ O	20.0mg
MnSO ₄ ·H ₂ O	20.0mg
NaCl	20.0mg

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Add standard solutions and test solutions to each tube. Bring volume of each tube to 10.0mL. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the microbiological assaying of riboflavin using *Lactobacillus casei* as the test organism.

Riboflavin Medium**Composition per liter:**

Glucose	20.0g
Yeast extract	10.0g
Peptone	10.0g
Riboflavin	0.02g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

**Rimler-Shotts Medium
(RS Medium)****Composition per liter:**

Agar	13.5g
Na ₂ S ₂ O ₃ ·5H ₂ O	6.8g
L-Ornithine·HCl	6.5g
NaCl	5.0g
L-Lysine·HCl	5.0g
Maltose	3.5g
Yeast extract	3.0g
Sodium deoxycholate	1.0g
Ferric ammonium citrate	0.8g
L-Cysteine·HCl	0.3g
Bromthymol Blue	0.03g
Novobiocin solution	10.0mL

pH 7.0 ± 0.2 at 25°C

Novobiocin Solution:**Composition per 10.0mL:**

Novobiocin	5.0mg

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile components. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and presumptive identification of *Aeromonas hydrophila* and other Gram-negative bacteria based on their ability to decarboxylate lysine and ornithine, ferment maltose, and produce H₂S. Maltose-fermenting bacteria appear as yellow colonies. Bacteria that produce lysine or ornithine decarboxylase turn the medium greenish-yellow to yellow. Bacteria that produce H₂S appear as colonies with black centers.

**Rippey-Cabelli Agar
(RC Agar)****Composition per liter:**

Agar	15.0g
Meat peptone	5.0g
Trehalose	5.0g
NaCl	3.0g
KCl	2.0g
Yeast extract	2.0g
Bromthymol Blue	0.44g
MgSO ₄ ·7H ₂ O	0.2g
FeCl ₃ ·6H ₂ O	0.1g
Sodium deoxycholate	0.1g
Ampicillin solution	10.0mL
Ethanol	10.0mL

pH 8.0 ± 0.2 at 25°C

Ampicillin Solution:**Composition per 10.0mL:**

Ampicillin	0.02g

Preparation of Ampicillin Solution: Add ampicillin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except sodium deoxycholate, ampicillin solution, and ethanol—to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sodium deoxycholate, 10.0mL of sterile ampicillin solution, and 10.0mL of ethanol. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and differentiation of *Aeromonas* species and *Plesiomonas* species from water samples using the membrane filter method. This medium differentiates bacteria on the basis of trehalose fermentation. Bacteria that ferment trehalose turn the medium yellow.

Rogosa Agar**Composition per liter:**

Sodium acetate	25.0g
Agar	20.0g
Glucose	20.0g
Pancreatic digest of casein	10.0g
KH ₂ PO ₄	6.0g
Yeast extract	5.0g
Ammonium citrate	2.0g
Sorbitan monooleate	1.0g
MgSO ₄ ·7H ₂ O	0.575g
MnSO ₄ ·H ₂ O	0.12g

FeSO ₄ ·7H ₂ O	0.4mg
Acetic acid, glacial.....	1.32mL
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and enumeration of lactobacilli, especially from dairy products.

Rogosa Broth, Modified

Composition per 1005.0mL:

Glucose	20.0g
Trypticase.....	10.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	3.0g
KH ₂ PO ₄	3.0g
Tryptose	3.0g
Ammonium citrate.....	2.0g
Sodium acetate.....	1.0g
Tween 80.....	1.0g
L-Cysteine	0.2g
Salt solution	5.0mL

pH 6.8 ± 0.2 at 25°C

Salt Solution:

Composition per 100.0mL:

MgSO ₄ ·7H ₂ O	11.50g
MnSO ₄	2.40g
FeSO ₄ ·7H ₂ O	1.68g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except salt solution, to distilled/deionized water and bring volume to 995.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 5.0mL of sterile salt solution. Adjust pH to 6.8. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Lactobacillus* species.

Rogosa SL Agar

(Rogosa Selective *Lactobacillus* Agar)

Composition per liter:

Agar	15.0g
Sodium acetate.....	15.0g

Glucose	10.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	6.0g
Yeast extract.....	5.0g
Arabinose.....	5.0g
Sucrose.....	5.0g
Ammonium citrate.....	2.0g
Sorbitan monooleate.....	1.0g
MgSO ₄ ·7H ₂ O	0.57g
MnSO ₄ ·7H ₂ O	0.12g
FeSO ₄ ·H ₂ O.....	0.03g
Acetic acid, glacial.....	1.32mL
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except glacial acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation, cultivation, and enumeration of lactobacilli, especially from dairy products.

Rogosa SL Broth

(Rogosa Selective *Lactobacillus* Broth)

Composition per liter:

Sodium acetate.....	15.0g
Glucose	10.0g
Pancreatic digest of casein.....	10.0g
K ₂ HPO ₄	6.0g
Yeast extract.....	5.0g
Arabinose.....	5.0g
Sucrose.....	5.0g
Ammonium citrate.....	2.0g
Sorbitan monooleate.....	1.0g
MgSO ₄ ·7H ₂ O	0.57g
MnSO ₄ ·7H ₂ O	0.12g
FeSO ₄ ·H ₂ O.....	0.03g
Acetic acid, glacial.....	1.32mL
pH 5.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except glacial acetic acid, to distilled/deionized water and bring volume to 998.7mL. Mix thoroughly. Gently heat and bring to boiling. Add glacial acetic acid. Mix thoroughly. Gently heat while stirring and bring to 90°–100°C for 2–3 min. Do not autoclave. Aseptically distribute into sterile tubes.

Use: For the isolation, cultivation, and enumeration of lactobacilli, especially from dairy products.

Rose Bengal Chloramphenicol Agar**Composition per liter:**

Agar	15.0g
Glucose	10.0g
Papaic digest of soybean meal	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
Rose Bengal	0.05g
Chloramphenicol solution	10.0mL
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Chloramphenicol Solution:**Composition per 10.0mL:**

Chloramphenicol	0.1g

Preparation of Chloramphenicol Solution:

Add chloramphenicol to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except chloramphenicol solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°C. Aseptically add sterile chloramphenicol solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation, cultivation, and enumeration of yeasts and molds from foods.

Russell Double-Sugar Agar**Composition per liter:**

Agar	15.0g
Proteose peptone No. 3	12.0g
Lactose	10.0g
NaCl	5.0g
Beef extract	1.0g
Glucose	1.0g
Phenol Red	0.025g
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position.

Use: For the identification of Gram-negative enteric bacilli based on their fermentation of glucose and lactose. Bacteria that ferment both glucose and lactose produce a yellow slant and yellow butt. Bacteria that ferment glucose but do not ferment lactose produce a red slant and a yellow butt. Bacteria that fer-

ment neither glucose nor lactose produce an unchanged pink-orange color.

S. aureus* ID*Composition per liter:**

Proprietary

Source: This medium is available from bioMérieux.

Use: For the direct identification of *Staphylococcus aureus* and the selective isolation of staphylococci. Direct identification of *S. aureus* is based on the spontaneous green coloration of α-glucosidase-producing colonies.

S Broth**Composition per liter:**

Peptone	10.0g
Meat extract	2.4g
NaCl	2.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Bacillus cereus*.

SA Agar**Composition per liter:**

Agar	15.0g
Pancreatic digest of casein	10.0g
NaCl	5.0g
Starch, soluble	1.0g
Ampicillin	0.01g
Phenol Red	0.018g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components, except ampicillin, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add ampicillin. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, and differentiation, based on starch hydrolysis, of *Aeromonas hydrophila* from foods. After inoculation of plates and growth of cultures, starch hydrolysis is determined by flooding each plate with 5.0mL of Lugol's iodine solution.

**SA Agar, Modified
(Lachica's Medium)****Composition per liter:**

Beef heart, solids from infusion	500.0g

Agar	15.0g
Tryptose	10.0g
NaCl	5.0g
Amylose azure	3.0g
Ampicillin	0.01mg

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation and cultivation of *Aeromonas hydrophila* from foods. *Aeromonas hydrophila* appears as colonies surrounded by a light halo on a light blue background.

Sabouraud Agar

Composition per liter:

Neopeptone	30.0g
Agar	20.0g

pH 5.6 ± 0.2 at 25°C

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds.

Sabouraud Agar, Diluted 1/10

Composition per liter:

Agar	20.0g
Glucose	4.0g
KH ₂ PO ₄	1.5g
MgSO ₄ ·7H ₂ O	1.0g
NaNO ₃	1.0g
Peptone	1.0g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.8. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of fungi and heterotrophic bacteria.

Sabouraud Agar, Modified

Composition per liter:

Agar	20.0g
Glucose	20.0g
Neopeptone	10.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts, molds, and aciduric bacteria.

Sabouraud Dextrose Agar (BAM M133)

Composition per liter:

Neopeptone	30.0g
Agar	20.0g

pH 5.6 ± 0.2 at 25°C

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of yeasts and molds.

Sabouraud Dextrose Broth (BAM M133)

Composition per liter:

Neopeptone	30.0g
Agar	20.0g

pH 5.6 ± 0.2 at 25°C

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of yeasts and molds.

Sabouraud Maltose Agar

Composition per liter:

Maltose	40.0g
Agar	15.0g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g

pH 5.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Avoid overheating. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of fungi.

Sabouraud Maltose Broth

Composition per liter:

Maltose.....	40.0g
Neopeptone.....	10.0g

pH 5.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Avoid overheating.

Use: For the cultivation of a variety of fungi.

Sabouraud Medium, Fluid

Composition per liter:

Glucose.....	20.0g
Pancreatic digest of casein.....	5.0g
Peptamin.....	5.0g

pH 5.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Avoid overheating.

Use: For the isolation and cultivation of yeasts and molds.

Saccharolytic Clostridia Medium

Composition per liter:

Pancreatic digest of casein.....	10.0g
Yeast extract.....	6.0g
Sodium thioglycolate.....	0.5g
Carbohydrate solution.....	100.0mL
Potassium phosphate (1M solution, pH 7.5).....	30.0mL
MgSO ₄ (1M solution).....	1.0mL
Solution M.....	0.5mL
FeSO ₄ solution.....	0.2mL

pH 7.0–7.2 at 25°C

Carbohydrate Solution:

Composition per 100.0mL:

Glucose or sucrose.....	20.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Solution M:

Composition per liter:

CaCl ₂	3.33g
MnCl ₂ ·4H ₂ O.....	1.98g
Na ₂ MoO ₄ ·2H ₂ O.....	1.21g
CoCl ₂ ·6H ₂ O.....	1.19g

Preparation of Solution M: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

FeSO₄·7H₂O Solution:

Composition per 10.0mL:

FeSO ₄ ·7H ₂ O.....	55.6g
H ₂ SO ₄ (0.1M solution).....	10.0mL

Preparation of FeSO₄·7H₂O Solution: Add the FeSO₄·7H₂O to 10.0mL of H₂SO₄ solution. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile carbohydrate solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the cultivation of saccharolytic *Clostridium* species.

Saccharomyces Medium

Composition per liter:

Glucose.....	20.0g
Peptone.....	20.0g
Agar.....	15.0g
Yeast extract.....	10.0g
Yeast nitrogen base without amino acids.....	6.7g
(NH ₄) ₂ SO ₄	5.0g
Proline.....	4.0mg

Preparation of Medium: Add glucose, yeast nitrogen base without amino acids, and proline to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Filter sterilize. Add peptone, yeast extract, (NH₄)₂SO₄, and agar to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically combine the two sterile solutions. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

Saccharomyces rouxii* Medium*Composition per liter:**

Glucose	400.0g
Agar	20.0g
Peptone.....	20.0g
Yeast extract.....	10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Zygosaccharomyces rouxii*.

***Salmonella* Chromogen Agar
(Rambach Equivalent Agar)****Composition per liter:**

Agar	15.0g
Peptone.....	5.0g
NaCl.....	5.0g
Yeast extract.....	2.0g
Meat extract	1.0g
Sodium deoxycholate.....	1.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: A differential diagnostic agar for the detection of *Salmonella* in food, including the isolation and enumeration of *Salmonella* from bivalves.

***Salmonella* Chromogenic Agar Base
(OSCM)****Composition per liter:**

Chromogenic mix	28.0g
Agar	12.0g
Special Peptone.....	10.0g
<i>Salmonella</i> selective supplement.....	10.0mL
pH 7.2 ± 0.2 at 25°C	

Salmonella* Selective Supplement:*Composition per 10.0mL:**

Cefulodin	12.0mg
Novobiocin.....	5.0mg

Preparation of *Salmonella* Selective Supplement: Add components to distilled/deionized water

and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to a boil to dissolve the components. Do not hold at boiling. Do not autoclave. Cool to 50°C. Aseptically add 10.0mL *Salmonella* selective supplement. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the identification of *Salmonella* spp. based upon the ability to utilize a specific chromogenic substrate which allows differentiation of *Salmonella* spp. from other members of the *Enterobacteriaceae*. The medium combines two chromogens for the detection of *Salmonella* spp., 5-Bromo-6-Chloro-3-Indolyl caprylate (Magenta-caprylate) and 5-Bromo-4-chloro-3-indolyl β-D-galactopyranoside (X-gal). X-gal is a substrate for the enzyme β-D-galactosidase. Hydrolysis of the chromogen, Mag-caprylate, by lactose negative *Salmonella* species results in magenta colonies. The medium contains bile salts to inhibit the growth of Gram-positive organisms and the addition of the *Salmonella* selective supplement increases the selectivity of the medium by inhibiting growth of *Proteus* spp. and *Pseudomonas* spp.

Salmonella* Medium*Composition per liter:**

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli* and *Salmonella choleraesuis*.

Salmonella* Rapid Test Elective Medium*Composition per liter:**

Tryptone.....	10.0g
Na ₂ HPO ₄	9.0g
Sodium chloride.....	5.0g
Casein	5.0g
KH ₂ PO ₄	1.5g
Malachite Green.....	0.0025g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix

thoroughly. Autoclave for 15 min at 15 psi pressure—121°C.

Use: For the Oxoid *Salmonella* Rapid Test which is for the presumptive detection of motile *Salmonella* spp. in foods.

***Salmonella* Rapid Test Elective Medium, 2X**

Composition per liter:

Tryptone.....	20.0g
Na ₂ HPO ₄	18.0g
Sodium chloride.....	10.0g
KH ₂ PO ₄	3.0g
Casein.....	10.0g
Malachite Green.....	0.005g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure—121°C.

Use: Use as described in the Oxoid *Salmonella* Rapid Test which is for the presumptive detection of motile *Salmonella* spp. in foods and environmental samples.

***Salmonella Shigella* Agar (SS Agar)**

Composition per liter:

Agar.....	13.5g
Lactose.....	10.0g
Bile salts.....	8.5g
Na ₂ S ₂ O ₃	8.5g
Sodium citrate.....	8.5g
Beef extract.....	5.0g
Pancreatic digest of casein.....	2.5g
Peptic digest of animal tissue.....	2.5g
Ferric citrate.....	1.0g
Neutral Red.....	0.025g
Brilliant Green.....	0.33mg

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes in 20.0mL volumes. Allow the surface of the plates to dry before inoculation.

Use: For the selective isolation and differentiation of pathogenic enteric bacilli, especially those belonging to the genus *Salmonella*. This medium is not recommended for the primary isolation of *Shigella* species.

Lactose-fermenting bacteria such as *Escherichia coli* or *Klebsiella pneumoniae* appear as small pink or red colonies. Lactose-nonfermenting bacteria—such as *Salmonella* species, *Proteus* species, and *Shigella* species—appear as colorless colonies. Production of H₂S by *Salmonella* species turns the center of the colonies black.

***Salmonella Shigella* Agar, Modified (SS Agar, Modified)**

Composition per liter:

Agar.....	12.0g
Lactose.....	10.0g
Sodium citrate.....	10.0g
Na ₂ S ₂ O ₃	8.5g
Bile salts.....	5.5g
Beef extract.....	5.0g
Peptone.....	5.0g
Ferric citrate.....	1.0g
Neutral Red.....	0.025g
Brilliant Green.....	0.33mg

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes in 20.0mL volumes. Allow the surface of the plates to dry before inoculation.

Use: For the selective isolation and differentiation of pathogenic enteric bacilli, especially those belonging to the genus *Salmonella*. This medium provides better growth of *Shigella* species. Lactose-fermenting bacteria such as *Escherichia coli* or *Klebsiella pneumoniae* appear as small pink or red colonies. Lactose-nonfermenting bacteria—such as *Salmonella* species, *Proteus* species, and *Shigella* species—appear as colorless colonies. Production of H₂S by *Salmonella* species turns the center of the colonies black.

Salt Broth, Modified

Composition per liter:

NaCl.....	65.0g
Enzymatic digest of animal tissue.....	10.0g
Heart digest.....	10.0g
Glucose.....	1.0g
Bromcresol Purple.....	0.016g

pH 7.2 ± 0.2 at 25°C

Source: Available as a prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of the enterococcal group D streptococci from nonenterococcal group D streptococci based on salt tolerance.

Salt Colistin Broth

Composition per liter:

NaCl.....	20.0g
Peptone.....	10.0g
Yeast extract.....	3.0g
Colistin solution.....	10.0mL
pH 7.4 ± 0.2 at 25°C	

Colistin Solution:

Composition per 10.0mL:

Colistin methane sulfonate.....	500,000U

Preparation of Colistin Solution: Add colistin methane sulfonate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except colistin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat until dissolved. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add sterile colistin solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of halophilic *Vibrio* species.

Salt Meat Broth

Composition per liter:

NaCl.....	100.0g
Neutral ox-heart tissue.....	30.0g
Beef extract.....	10.0g
Peptone.....	10.0g
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available as tablets from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of staphylococci from specimens with a mixed flora such as fecal specimens, especially during the investigation of staphylococcal food poisoning.

Salt Polymyxin Broth (SPB)

Composition per liter:

NaCl.....	20.0g
Pancreatic digest of casein.....	10.0g
Yeast extract.....	3.0g
Polymyxin B.....	250,000U
pH 8.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.8. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–115°C.

Use: For the isolation and cultivation of *Vibrio* species from foods.

Salt Tolerance Medium

Composition per liter:

Beef heart, infusion from.....	500.0g
NaCl.....	65.0g
Tryptose.....	10.0g
Glucose.....	1.0g
Indicator solution.....	1.0mL
pH 7.4 ± 0.2 at 25°C	

Indicator Solution:

Composition per 100.0mL:

Bromcresol Purple.....	1.6g
Ethanol (95% solution).....	100.0mL

Preparation of Indicator Solution: Add Bromcresol Purple to ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of salt-tolerant *Streptococcus* species and other salt-tolerant Gram-positive cocci. For the differentiation of Gram-positive cocci based on salt tolerance.

Salt Tolerance Medium

Composition per liter:

NaCl.....	60.0g
Peptone.....	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Aeromonas* and *Plesiomonas* species based on salt tolerance.

Salt Tolerance Medium

Composition per liter:

Beef heart, solids from infusion.....	500.0g
NaCl.....	65.0g
Tryptose	10.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For testing the salt tolerance of a variety of microorganisms.

Salt Tolerance Medium, Gilardi

Composition per liter:

NaCl.....	65.0g
Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of salt-tolerant, nonfermenting Gram-negative bacteria. For the differentiation of nonfermenting Gram-negative bacteria based on salt tolerance.

Salt Tolerance Medium, Tatum

Composition per liter:

NaCl.....	65.0g
Peptone.....	5.0g
Yeast extract.....	2.0g
Beef extract.....	1.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of salt-tolerant, nonfermenting Gram-negative bacteria. For the differentiation of nonfermenting Gram-negative bacteria based on salt tolerance.

Sanfrancisco Medium

Composition per liter:

Rye bran or wheat bran.....	50.0g
Fresh baker's yeast	21.0g
Pancreatic digest of casein.....	10.0g
Fructose.....	7.0g
Glucose	7.0g
Maltose	7.0g
Yeast extract.....	7.0g
Diammonium citrate	5.0g
Sodium acetate·3H ₂ O.....	5.0g
KH ₂ PO ₄ ·3H ₂ O	2.5g
Meat extract	2.0g
Sodium gluconate	2.0g
Tween 80.....	1.0g
L-Cysteine·HCl	0.5g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
FeSO ₄ ·7H ₂ O.....	0.01g

pH 5.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.4. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus sanfrancisco*.

SBG Enrichment Broth (Selenite Brilliant Green Enrichment Broth)

Composition per liter:

D-Mannitol	5.0g
Peptone	5.0g
Yeast extract.....	5.0g
Na ₂ SeO ₃ ·5H ₂ O.....	4.0g
K ₂ HPO ₄	2.65g
KH ₂ PO ₄	1.02g
Sodium taurocholate	1.0g
Brilliant Green	5.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 5–10 min. Do not autoclave. Distribute into sterile tubes or flasks.

Use: For the selective isolation of *Salmonella* species, especially from eggs and egg products.

SBG Sulfa Enrichment**Composition per liter:**

D-Mannitol	5.0g
Peptone	5.0g
Yeast extract	5.0g
Na ₂ SeO ₃ ·5H ₂ O	4.0g
K ₂ HPO ₄	2.65g
KH ₂ PO ₄	1.02g
Sodium taurocholate	1.0g
Sodium sulfapyridine	0.5g
Brilliant Green	5.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 5–10 min. Do not autoclave. Distribute into sterile tubes or flasks.

Use: For the selective isolation of *Salmonella* species, especially from eggs and egg products.

SC Agar**Composition per liter:**

Agar	20.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine-HCl	0.4mg
Pyroxidine-HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid	2.0μg
Biotin	2.0μg
Glucose solution	50.0mL
Synthetic stock solution	30.0mL

Glucose Solution:**Composition per 100.0mL:**

Glucose	40.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Synthetic Stock Solution:**Composition per 300.0mL:**

L-Isoleucine	5.25g
L-Arginine	3.48g
L-Aspartic acid	2.66g
L-Leucine	2.62g
L-Methionine	1.49g
L-Threonine	1.19g
L-Valine	1.17g
L-Serine	1.05g
L-Lysine	0.91g
L-Phenylalanine	0.83g
L-Tryptophan	0.82g
L-Histidine	0.58g
<i>m</i> -Inositol	0.36g
Uracil	0.22g
L-Tyrosine	0.18g
Adenine	0.135g

Preparation of Synthetic Stock Solution: Add components to distilled/deionized water and bring volume to 300.0mL. Mix thoroughly. Filter sterilize. Store in dark at 25°C.

Preparation of Medium: Add components, except synthetic stock solution and glucose solution, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure—121°C. Cool to 50°–55°C. Aseptically add 30.0mL of sterile synthetic stock solution and 50.0mL of sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

SC Broth**Composition per liter:**

(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine-HCl	0.4mg
Pyroxidine-HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg

<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg
Glucose solution	50.0mL
Synthetic stock solution.....	30.0mL

Glucose Solution:**Composition per 100.0mL:**

Glucose	40.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Synthetic Stock Solution:**Composition per 300.0mL:**

L-Isoleucine.....	5.25g
L-Arginine	3.48g
L-Aspartic acid.....	2.66g
L-Leucine	2.62g
L-Methionine.....	1.49g
L-Threonine.....	1.19g
L-Valine	1.17g
L-Serine	1.05g
L-Lysine	0.91g
L-Phenylalanine	0.83g
L-Tryptophan.....	0.82g
L-Histidine	0.58g
<i>m</i> -inositol	0.36g
Uracil	0.22g
L-Tyrosine	0.18g
Adenine.....	0.135g

Preparation of Synthetic Stock Solution: Add components to distilled/deionized water and bring volume to 300.0mL. Mix thoroughly. Filter sterilize. Store in dark at 25°C.

Preparation of Medium: Add components, except synthetic stock solution and glucose solution, to distilled/deionized water and bring volume to 920.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 30.0mL of sterile synthetic stock solution and 50.0mL of sterile glucose solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Schaedler Agar**(Schaedler Anaerobic Agar)****Composition per liter:**

Agar	13.5g
Glucose	5.83g

Pancreatic digest of casein.....	5.7g
Proteose peptone No. 3	5.0g
Yeast extract.....	5.0g
Tris(hydroxymethyl)aminomethane buffer.....	3.0g
NaCl.....	1.65g
Papaic digest of soybean meal.....	1.0g
K ₂ HPO ₄	0.83g
L-Cystine	0.4g
Hemin	0.01g

pH 7.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and enumeration of anaerobic and aerobic microorganisms.

Schaedler Agar**Composition per liter:**

Agar	13.5g
Pancreatic digest of casein.....	8.2g
Glucose	5.8g
Yeast extract.....	5.0g
Tris(hydroxymethyl)aminomethane buffer.....	3.0g
Peptic digest of animal tissue	2.5g
NaCl.....	1.7g
Papaic digest of soybean meal.....	1.0g
K ₂ HPO ₄	0.8g
L-Cystine	0.4g
Hemin	0.01g

pH 7.6 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and enumeration of anaerobic and aerobic microorganisms.

Schleifer-Krämer Agar**(SK Agar)****Composition per liter:**

Agar	13.0g
Glycerol	10.0g
Sodium pyruvate.....	10.0g

Pancreatic digest of casein.....	10.0g
Beef extract.....	5.0g
Yeast extract.....	3.0g
Potassium isothiocyanate.....	2.25g
LiCl.....	2.0g
Na ₂ HPO ₄ ·2H ₂ O.....	0.9g
NaH ₂ PO ₄ ·H ₂ O.....	0.6g
Glycine.....	0.5g
NaN ₃ solution.....	10.0mL
pH 7.2 ± 0.2 at 25°C	

NaN₃ Solution:**Composition per 10.0mL:**

NaN ₃	0.045g

Preparation of NaN₃ Solution: Add NaN₃ to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except NaN₃ solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile NaN₃ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Staphylococcus* species.

Seawater Agar (SWA)

Composition per liter:

Agar.....	15.0g
Peptone.....	5.0g
Yeast extract.....	5.0g
Beef extract.....	3.0g
Seawater, synthetic.....	1.0L
pH 7.5 ± 0.2 at 25°C	

Seawater, Synthetic:**Composition per liter:**

NaCl.....	27.0g
MgSO ₄ ·7H ₂ O.....	7.0g
Tris(hydroxymethyl)aminomethane buffer.....	2.0g
KCl.....	0.6g
CaCl ₂	0.3g

Preparation of Seawater, Synthetic: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of halophilic microorganisms from foods, such as *Pseudomonas* species and *Vibrio* species from fish.

Seawater Agar (SWA)

Composition per liter:

Agar.....	15.0g
Peptone.....	5.0g
Yeast extract.....	5.0g
Beef extract.....	3.0g
Seawater, synthetic.....	1.0L
pH 7.5 ± 0.2 at 25°C	

Seawater, Synthetic:**Composition per liter:**

NaCl.....	24.0g
MgSO ₄ ·7H ₂ O.....	7.0g
MgCl ₂ ·6H ₂ O.....	5.3g
KCl.....	0.7g
CaCl ₂	0.1g

Preparation of Seawater, Synthetic: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.5.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of halophilic microorganisms from foods, such as *Pseudomonas* species and *Vibrio* species from fish.

Seawater Agar with Fetal Calf Serum

Composition per liter:

Agar.....	20.0g
Beef extract.....	10.0g
Peptone.....	10.0g
Seawater.....	750.0mL
Fetal calf serum.....	100.0mL
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components, except fetal calf serum, to tap water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile fetal calf serum warmed to 50°–55°C. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Aeromonas* species and *Vibrio salmonicida*.

Seawater Agar with Horse Blood

Composition per liter:

Agar	20.0g
Beef extract	10.0g
Peptone	10.0g
Seawater	750.0mL
Horse blood	100.0mL

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components, except horse blood, to tap water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile horse blood warmed to 50°–55°C. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Aeromonas* species.

Seawater Yeast Extract Broth, Modified

Composition per liter:

NaCl	23.4g
MgSO ₄ ·7H ₂ O	6.9g
Peptone	1.0g
Yeast extract	1.0g
KCl	0.75g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Proteus* species and *Vibrio* species.

Selection Agar I

Composition per liter:

Agar	25.0g
Yeast extract	1.0g
CuSO ₄	0.5mg
Wort solution	1.0L

Wort Solution:

Composition per 1.0L:

Malt extract	110.0g

Preparation of Wort Solution: Add malt extract to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective cultivation of *Saccharomyces cerevisiae*.

Selection Agar II

Composition per liter:

Sodium acetate	40.0g
Agar	25.0g
(NH ₄) ₂ SO ₄	5.0g
Maltose	2.5g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine·HCl	0.4mg
Pyroxidine·HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
p-Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid	2.0μg
Biotin	2.0μg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective cultivation of *Saccharomyces cerevisiae*.

Selenite Broth

(Selenite Broth, Lactose) (Selenite F Enrichment Medium) (Sodium Biselenite Medium)

(Sodium Hydrogen Selenite Medium)

Composition per liter:

Na ₂ HPO ₄	10.0g
Pancreatic digest of casein	5.0g
Lactose	4.0g
NaHSO ₃ ·5H ₂ O	4.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and a prepared medium from Oxoid.

Caution: Sodium biselenite is toxic and a potential teratogen and care must be taken to avoid inhalation of the powdered dye, contact with the skin, or ingestion, especially in pregnant laboratory workers.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Distribute into sterile tubes in 10.0mL volumes.

Use: For the isolation and enrichment of *Salmonella* species from clinical specimens and food products.

Selenite Broth Base, Mannitol

Composition per liter:

Na ₂ HPO ₄	10.0g
Peptone.....	5.0g
Mannitol.....	4.0g
NaHSeO ₃ ·5H ₂ O	4.0g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Caution: Sodium selenite is toxic and a potential teratogen and care must be taken to avoid inhalation of the powdered dye, contact with the skin, or ingestion, especially in pregnant laboratory workers.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat. Do not autoclave. Distribute into sterile tubes in 10.0mL volumes. Sterilize for 10 min at 0 psi pressure–100°C.

Use: For the isolation and cultivation of *Salmonella typhi* and *Salmonella paratyphi*.

Selenite Cystine Broth (BAM M134 Medium 1)

Composition per liter:

Na ₂ HPO ₄	10.0g
Pancreatic digest of casein.....	5.0g
Lactose.....	4.0g
Na ₂ SeO ₃ ·5H ₂ O.....	4.0g
L-Cystine	0.01g
pH 7.0 ± 0.2 at 25°C	

Caution: Sodium selenite is toxic and a potential teratogen and care must be taken to avoid inhalation of the powdered dye, contact with the skin, or ingestion, especially in pregnant laboratory workers.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat. Do not autoclave. Distribute into sterile tubes in 10.0mL volumes. Sterilize for 10 min at 0 psi pressure–100°C in flowing steam.

Use: For the isolation and cultivation of *Salmonella* species from feces, dairy products, and other specimens.

Selenite Cystine Broth (BAM M134)

Composition per liter:

Na ₂ HPO ₄	5.5g
Polypeptone.....	5.0g
KH ₂ PO ₄	4.5g
Lactose.....	4.0g
Na ₂ SeO ₃ ·5H ₂ O.....	4.0g
L-Cystine	0.01g
pH 7.0 ± 0.2 at 25°C	

Caution: Sodium selenite is toxic and a potential teratogen and care must be taken to avoid inhalation of the powdered dye, contact with the skin, or ingestion, especially in pregnant laboratory workers.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat. Do not autoclave. Distribute into sterile tubes in 10.0mL volumes. Sterilize for 10 min at 0 psi pressure–100°C in flowing steam.

Use: For the isolation and cultivation of *Salmonella* species from feces, dairy products, and other specimens.

Selenite F Broth

Composition per liter:

KH ₂ PO ₄	7.0g
Pancreatic digest of casein.....	5.0g
Lactose.....	4.0g
Na ₂ SeO ₃ ·5H ₂ O.....	4.0g
Na ₂ HPO ₄	3.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Caution: Sodium selenite is toxic and a potential teratogen and care must be taken to avoid inhalation of the powdered dye, contact with the skin, or ingestion, especially in pregnant laboratory workers.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat. Do not autoclave. Distribute into sterile tubes in 10.0mL volumes. Sterilize for 30 min at 0 psi pressure–100°C.

Use: For the isolation and cultivation of *Salmonella* species from feces, dairy products, and other specimens.

Semisolid Brucella Broth

Composition per liter:

Peptone.....	10.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g
Yeast extract.....	2.0g

Agar	1.6g
D-Glucose.....	1.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Campylobacter* species.

Semi-Solid Medium, Modified (BAM M30c)

Composition per liter:

Peptone.....	10.0g
Lactalbumin hydrolysate.....	5.0g
Yeast extract.....	5.0g
NaCl.....	5.0g
Agar	1.8g
α -Ketoglutaric acid.....	1.0g
Na-pyruvate	0.5g
$\text{Na}_2\text{S}_2\text{O}_5$	0.5g
NaCO_3	0.6g
Na-citrate	0.1g
Hemin.....	0.01g

pH 7.4 $\pm$ 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the storage and preservation of *Campylobacter* spp.

Semisolid Medium, Modified with Glycine and Neutral Red (BAM M30d)

Composition per liter:

Glycine.....	10.0g
Peptone.....	10.0g
Lactalbumin hydrolysate.....	5.0g
Yeast extract.....	5.0g
NaCl.....	5.0g
Agar	1.8g
α -Ketoglutaric acid.....	1.0g
NaCO_3	0.6g
Na-pyruvate	0.5g
$\text{Na}_2\text{S}_2\text{O}_5$	0.5g
Hemin.....	0.01g
Neutral Red solution	10.0mL

pH 7.4 $\pm$ 0.2 at 25°C

Neutral Red Solution:

Composition per 100.0mL:

Neutral Red.....	0.2g
Ethanol	10.0mL

Preparation of Neutral Red Solution: Add 0.2g Neutral Red to 10.0mL ethanol. Mix thoroughly. Bring volume to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the biochemical identification of *Campylobacter* spp.

Semisolid Medium, Modified with NaCl and Neutral Red (BAM M30d)

Composition per liter:

NaCl.....	35.0g
Peptone	10.0g
Lactalbumin hydrolysate	5.0g
Yeast extract.....	5.0g
Agar	1.8g
α -Ketoglutaric acid.....	1.0g
NaCO_3	0.6g
Na-pyruvate	0.5g
$\text{Na}_2\text{S}_2\text{O}_5$	0.5g
Hemin	0.01g
Neutral Red solution	10.0mL

pH 7.4 $\pm$ 0.2 at 25°C

Neutral Red Solution:

Composition per 100.0mL:

Neutral Red.....	0.2g
Ethanol.....	10.0mL

Preparation of Neutral Red Solution: Add 0.2g Neutral Red to 10.0mL ethanol. Mix thoroughly. Bring volume to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the biochemical identification of *Campylobacter* spp.

Semisolid Medium, Modified with Cysteine and Neutral Red (BAM M30d)

Composition per liter:

Peptone	10.0g
Lactalbumin hydrolysate	5.0g

Yeast extract.....	5.0g
NaCl.....	5.0g
Agar.....	1.8g
α -Ketoglutaric acid.....	1.0g
NaCO ₃	0.6g
Na-pyruvate.....	0.5g
Na ₂ S ₂ O ₅	0.5g
L-Cysteine-HCl·H ₂ O.....	0.2g
Hemin.....	0.01g
Neutral Red solution.....	10.0mL
pH 7.4 ± 0.2 at 25°C	

Neutral Red Solution:**Composition per 100.0mL:**

Neutral Red.....	0.2g
Ethanol.....	10.0mL

Preparation of Neutral Red Solution: Add 0.2g Neutral Red to 10.0mL ethanol. Mix thoroughly. Bring volume to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the biochemical identification of *Campylobacter* spp.

**Semisolid Medium, Modified
with Nitrate and without Neutral Red
(BAM M30d)**

Composition per liter:

KNO ₃	10.0g
Peptone.....	10.0g
Lactalbumin hydrolysate.....	5.0g
Yeast extract.....	5.0g
NaCl.....	5.0g
Agar.....	1.8g
α -Ketoglutaric acid.....	1.0g
NaCO ₃	0.6g
Na-pyruvate.....	0.5g
Na ₂ S ₂ O ₅	0.5g
Hemin.....	0.01g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool to 50°C. Adjust pH to 7.4. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the biochemical identification of *Campylobacter* spp.

Semisolid Medium for Motility**Composition per liter:**

Biosate.....	5.0g
Polypeptone.....	5.0g
NaCl.....	5.0g
Agar.....	4.0g
Myosate.....	1.5g
Triphenyltetrazolium chloride solution.....	2.5mL
pH 6.9–7.1 at 25°C	

Triphenyltetrazolium Chloride Solution:**Composition per 10.0mL:**

Triphenyltetrazolium chloride.....	0.1g
Ethanol (95% solution).....	10.0mL

Preparation of Triphenyltetrazolium Chloride Solution: Add triphenyltetrazolium chloride to 10.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components, except triphenyltetrazolium chloride solution, to distilled/deionized water and bring volume to 997.5mL. Mix thoroughly. Gently heat and bring to boiling. Add 2.5mL of triphenyltetrazolium chloride solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of bacteria based on motility.

**Serum Glucose Agar
(Serum Dextrose Agar)
(ATCC Medium 287)**

Composition per 1060.0mL:

Agar.....	15.0g
Peptone.....	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g
Serum-glucose solution.....	60.0mL
pH 7.3 ± 0.2 at 25°C	

Serum-Glucose Solution:**Composition per 60.0mL:**

D-Glucose.....	10.0g
Serum (inactivated at 56°C, 30 min).....	50.0mL

Preparation of Serum-Glucose Solution: Add glucose to 50.0mL of heat-inactivated serum. Horse serum or ox serum may be used. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except serum-glucose solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 10 psi pressure–115°C. Cool to 50°C. Aseptically add 60.0mL of sterile serum-glucose solution. Mix thoroughly. Pour into sterile Petri dishes or dis-

tribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation and maintenance of *Brucella* species.

Serum Glucose Agar, Farrell Modified

Composition per 1086.9mL:

Agar	15.0g
Peptone.....	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g
Serum-glucose solution.....	60.0mL
Bacitracin solution	12.5mL
Cycloheximide solution	10.0mL
Nystatin solution.....	2.0mL
Polymyxin B solution	1.0mL
Nalidixic acid solution.....	1.0mL
Vancomycin solution.....	0.4mL

pH 7.3 ± 0.2 at 25°C

Serum-Glucose Solution:

Composition per 60.0mL:

D-Glucose	10.0g
Serum (inactivated at 56°C , 30 min)	50.0mL

Preparation of Serum-Glucose Solution: Add glucose to 50.0mL of heat-inactivated serum. Horse serum or ox serum may be used. Mix thoroughly. Filter sterilize.

Bacitracin Solution:

Composition per 12.5mL:

Bacitracin.....	25,000U

Preparation of Bacitracin Solution: Add Bacitracin to distilled/deionized water and bring volume to 12.5mL. Mix thoroughly. Filter sterilize.

Cycloheximide Solution:

Composition per 100.0mL:

Cycloheximide.....	1.0g
Acetone	5.0mL

Preparation of Cycloheximide Solution: Add cycloheximide to 5.0mL of acetone. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water. Mix thoroughly. Filter sterilize.

Nystatin Solution:

Composition per 5.0mL:

Nystatin	250,000U

Preparation of Nystatin Solution: Add nystatin to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly. Filter sterilize.

Polymyxin B Solution:

Composition per 2.0mL:

Polymyxin B	10,000U

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 2.0mL. Mix thoroughly. Filter sterilize.

Nalidixic Acid Solution:

Composition per 2.0mL:

Nalidixic acid.....	0.1g
NaOH (0.5N solution).....	2.0mL

Preparation of Nalidixic Acid Solution: Add nalidixic acid to 2.0mL of NaOH solution. Mix thoroughly. Immediately before use, add 1.0mL of this stock solution to 9.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize.

Vancomycin Solution:

Composition per 1.0mL:

Vancomycin	0.05g

Preparation of Vancomycin Solution: Add vancomycin to distilled/deionized water and bring volume to 1.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except serum-glucose solution, bacitracin solution, cycloheximide solution, nystatin solution, polymyxin B solution, nalidixic acid solution, and vancomycin solution—to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 10 psi pressure— 115°C . Cool to 50°C . Aseptically add 60.0mL of sterile serum-glucose solution, 12.5mL of sterile bacitracin solution, 10.0mL of sterile cycloheximide solution, 2.0mL of sterile nystatin solution, 1.0mL of sterile polymyxin B solution, 1.0mL of sterile nalidixic acid solution, and 0.4mL of sterile vancomycin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the selective isolation and cultivation of *Brucella* species.

Serum Potato Infusion Agar

Composition per 1120.0mL:

Agar	15.0g
Peptone	10.0g
Meat extract	5.0g
NaCl.....	5.0g
Potato infusion.....	1.0L
Horse serum, heat inactivated.....	100.0mL
Glycerol	20.0mL

pH 6.8 ± 0.2 at 25°C

Potato Infusion:

Composition per 10.0mL:

Potatoes.....	250.0g

Preparation of Potato Infusion: Add peeled, thinly sliced potatoes to 1.0L of distilled/deionized

water. Infuse overnight at 60°C. Filter through Whatman #1 filter paper. Bring volume to 1.0L with distilled/deionized water.

Preparation of Medium: Combine components, except horse serum. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of sterile horse serum. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Brucella* species.

Seven H11 Agar (Selective 7H11 Agar)

Composition per 1010.0mL:

Agar	13.5g
KH ₂ PO ₄	1.5g
Na ₂ HPO ₄	1.5g
Pancreatic digest of casein	1.0g
NaCl.....	0.85g
Monosodium glutamate	0.5g
(NH ₄) ₂ SO ₄	0.5g
Sodium citrate	0.4g
MgSO ₄ ·7H ₂ O	0.05g
Ferric ammonium citrate.....	0.04g
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine	1.0mg
ZnSO ₄ ·7H ₂ O	1.0mg
Biotin	0.5mg
CaCl ₂ ·2H ₂ O.....	0.5mg
Malachite Green.....	0.25mg
Glycerol	5.0mL
Middlebrook OADC enrichment	100.0mL
Antibiotic inhibitor	10.0mL
pH 6.6 ± 0.2 at 25°C	

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Middlebrook OADC Enrichment:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl.....	0.85g
Catalase.....	3.0mg
Oleic acid	0.06mL

Preparation of Middlebrook OADC Enrichment: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Antibiotic Inhibitor:

Composition per 10.0mL:

Carbenicillin.....	0.05g
Trimethoprim lactate.....	0.02g

Amphotericin B	0.01g
Polymyxin B	200,000U

Preparation of Antibiotic Inhibitor: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add glycerol to 900.0mL of distilled/deionized water. Mix thoroughly. Add remaining components, except Middlebrook OADC enrichment and antibiotic inhibitor. Mix thoroughly. Gently heat. Do not boil. Autoclave for 10 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile Middlebrook OADC enrichment and 10.0mL of sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Mycobacterium* species from specimens with a mixed flora.

Seven-Hour Fecal Coliform Agar (Seven-Hour FC Agar)

(m-Seven-Hour Fecal Coliform Agar)

Composition per liter:

Agar	15.0g
Lactose	10.0g
NaCl.....	7.5g
D-Mannitol	5.0g
Proteose peptone No. 3	5.0g
Yeast extract.....	3.0g
Bromocresol Purple	0.35g
Phenol Red.....	0.3g
Sodium lauryl sulfate	0.2g
Sodium deoxycholate.....	0.1g
pH 7.3 ± 0.1 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 5 min. Cool to 55°–60°C. Adjust pH to 7.3 with 0.1N NaOH. Cool to 45°–50°C. Pour into sterile Petri dishes with tight-fitting lids in 5.0mL volumes. Store at 2°–10°C.

Use: For the rapid estimation of the bacteriological quality of water using the membrane filter method.

SF Broth (*Streptococcus faecalis* Broth)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Glucose	5.0g
NaCl.....	5.0g
K ₂ HPO ₄	4.0g
KH ₂ PO ₄	1.5g

NaN ₃	0.5g
Bromcresol Purple	0.032g
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of group D enterococci (*Streptococcus faecalis* and *Streptococcus faecium*) from group D nonenterococci and from other *Streptococcus* species. Group D enterococci turn the medium turbid and yellow-brown.

SFP Agar

(Shahidi-Ferguson Perfringens Agar)

Composition per 2020.0mL:

Basal layer	1010.0mL
Cover layer	1010.0mL

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Basal Layer:

Composition per 1010.0mL:

Agar	20.0g
Tryptose	15.0g
Papaic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
NaHSO ₃	1.0g
Egg yolk emulsion, 50%	100.0mL
Antibiotic inhibitor	10.0mL
pH 7.6 ± 0.2 at 25°C	

Egg Yolk Emulsion, 50%:

Composition per 100.0mL:

Chicken egg yolks	11
Whole chicken egg	1
NaCl (0.9% solution)	50.0mL

Preparation of Egg Yolk Emulsion, 50%:

Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg. Measure 50.0mL of egg yolk emulsion and add to 50.0mL of 0.9% NaCl solution. Mix thoroughly. Filter sterilize. Warm to 45°–50°C.

Antibiotic Inhibitor:

Composition per 10.0mL:

Kanamycin	0.012g
Polymyxin B sulfate	30,000U

Preparation of Antibiotic Inhibitor: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Basal Layer: Add components—except egg yolk emulsion, 50%, and antibiotic inhibitor—to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile egg yolk emulsion, 50%, and antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes in 10.0mL volumes.

Cover Layer:

Composition per 1010.0mL:

Agar	20.0g
Tryptose	15.0g
Papaic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
NaHSO ₃	1.0g
Antibiotic inhibitor	10.0mL
pH 7.6 ± 0.2 at 25°C	

Preparation of Cover Layer: Add components—except antibiotic inhibitor—to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic inhibitor. Mix thoroughly.

Preparation of Medium: Prepare and dispense basal layer into sterile Petri dishes in 10.0mL volumes. Incubate overnight to dry plates and test for sterility. Inoculate plates using 0.1mL volume. Spread inoculum over surface of agar. Aseptically add 10.0mL of cover layer to each plate. Incubate at 37°C under 90% N₂ + 10% CO₂.

Use: For the isolation and enumeration of *Clostridium perfringens* from foods. *Clostridium perfringens* appears as black colonies surrounded by a precipitate.

Sheep Blood Agar (BAM M135)

Composition per liter:

Proteose peptone	15.0g
Agar	12.0g
Liver digest	2.5g
Yeast extract	5.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–46°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of streptococci and other fastidious microorganisms.

Sheep Blood Agar

Composition per liter:

Tryptone	14.0g
Agar	12.0g
NaCl	5.0g
Peptone, neutralized	4.5g
Yeast extract	4.5g
Sheep blood, defibrinated	70.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 930.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 70.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For the isolation, cultivation, and detection of hemolytic activity of streptococci and other fastidious microorganisms. Specifically formulated to give maximum recovery and improved hemolytic reactions with sheep blood.

Shigella Broth (BAM M136)

Composition per liter:

Pancreatic digest of casein	20.0g
NaCl	5.0g
K ₂ HPO ₄	2.0g
KH ₂ PO ₄	2.0g
Glucose	1.0g
Novobiocin solution	11.1mL
Tween 80	1.5mL
pH 7.0 ± 0.2 at 25°C	

Novobiocin Solution:

Composition per liter:

Novobiocin	0.05g

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 988.9mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile novobiocin solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the isolation and cultivation of *Shigella* species from food.

SI Agar

Composition per liter:

Peptone	15.6g
Agar	12.0g
NaCl	5.6g
Yeast extract	2.8g
D-Glucose	1.0g
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

Sierra Medium

Composition per liter:

Agar	15.0g
Peptone	10.0g
NaCl	5.0g
CaCl ₂ ·H ₂ O	0.1g
Tween 80	10.0mL
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components, except Tween 80, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Separately autoclave Tween 80 for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile Tween 80. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the differentiation of bacteria based on lipase activity. Bacteria with lipase activity form colonies surrounded by a white precipitate.

SIM Medium

Composition per liter:

Peptone.....	30.0g
Agar	3.0g
Beef extract.....	3.0g
Peptonized iron	0.2g
Na ₂ S ₂ O ₃ ·5H ₂ O	0.025g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 15.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the differentiation of members of the *Enterobacteriaceae* based on H₂S production, indole production, and motility.

SIM Motility Medium (BAM M137)

Composition per liter:

Pancreatic digest of casein.....	20.0g
Peptic digest of animal tissue.....	6.1g
Agar	3.5g
Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O	0.2g
Na ₂ S ₂ O ₃ ·5H ₂ O	0.2g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 15.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the differentiation of members of the *Enterobacteriaceae* based on H₂S production, indole production, and motility.

Simmons' Citrate Agar (Citrate Agar) (BAM M138)

Composition per liter:

Agar	15.0g
NaCl.....	5.0g
Sodium citrate	2.0g
K ₂ HPO ₄	1.0g
(NH ₄)H ₂ PO ₄	1.0g

MgSO ₄ ·7H ₂ O	0.2g
Bromthymol Blue	0.08g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of Gram-negative bacteria on the basis of citrate utilization. Bacteria that can utilize citrate as sole carbon source turn the medium blue.

Single-Layer Agar

Composition per 1050.0mL:

Basal medium	1.0L
Tributylin substrate.....	50.0g
pH 6.8 ± 0.2 at 25°C	

Basal Medium:

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g

Preparation of Basal Medium: Add agar to 1.0L of distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Tributylin Substrate:

Composition:

Tributylin substrate.....	50.0g

Preparation of Tributyrin Substrate: Remove free fatty acids in the tributyrin substrate by dissolving 50.0g in 500.0mL of petroleum ether. Pass the solution through an activated alumina column. Remove the petroleum ether by evaporation on a steam table under 100% N₂. Autoclave for 30 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Aseptically combine 1.0L of sterile basal medium with 50.0g of sterile tributyrin substrate in a warm, sterile blender container. Blend for 1 min until homogenized. Rapidly pour into sterile Petri dishes in 7.0mL volumes. Dry the surface of the plates by partially opening the lids in a laminar flow hood for 15 min.

Use: For the isolation, cultivation, and identification of lipolytic microorganisms from food.

Single-Layer Agar**Composition** per 1050.0mL:

Basal medium	1.0L
Fat substrate	50.0g
pH 6.8 ± 0.2 at 25°C	

Basal Medium:**Composition** per liter:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g
Victoria Blue B solution	200.0mL

Preparation of Basal Medium: Add agar to 800.0mL of distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 200.0mL of Victoria Blue B solution. Mix thoroughly.

Victoria Blue B Solution:**Composition** per 200.0mL:

Victoria Blue B	0.12g

Preparation of Victoria Blue B Solution: Add the Victoria Blue B to 200.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize. Warm to 50°C.

Fat Substrate:**Composition:**

Fat substrate	50.0g

Preparation of Fat Substrate: Corn oil, soybean oil, any cooking oil, lard, tallow, or triglycerides that do not contain antioxidants or other inhibitory substances may be used. Remove free fatty acids in the fat substrate by dissolving 50.0g of fat substrate in 500.0mL of petroleum ether. Pass the solution through an activated alumina column. Remove the petroleum ether by evaporation on a steam table under 100% N₂. Autoclave for 30 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Aseptically combine 1.0L of sterile basal medium with 50.0g of sterile fat substrate in a warm, sterile blender container. Blend for 1 min until homogenized. Rapidly pour into sterile Petri dishes in 7.0mL volumes. Dry the surface of the plates by partially opening the lids in a laminar flow hood for 15 min.

Use: For the isolation, cultivation, and identification of lipolytic microorganisms from food.

Skim Milk Agar**Composition** per 1100.0mL:

Agar	15.0g
Pancreatic digest of casein	5.0g
Yeast extract	2.5g

Glucose	1.0g
Skim milk solution	100.0mL
pH 7.0 ± 0.1 at 25°C	

Preparation of Skim Milk Solution: Add skim milk solids to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components, except skim milk solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 100.0mL of cooled, sterile skim milk solution. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and differentiation of bacteria based on proteolytic activity.

Skirrow *Brucella* Medium**Composition** per liter:

Blood agar base No. 2	940.0mL
Horse blood, lysed defibrinated	50.0mL
Antibiotic solution	10.0mL
pH 7.4 ± 0.2 at 25°C	

Blood Agar Base No. 2**Composition** per 940.0mL:

Proteose peptone	15.0g
Agar	12.0g
NaCl	5.0g
Yeast extract	5.0g
Liver digest	2.5g
pH 7.4 ± 0.2 at 25°C	

Preparation of Blood Agar Base No. 2: Add components to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Solution:**Composition** per 10.0mL:

Vancomycin	0.01g
Trimethoprim	5.0mg
Polymyxin B	2500U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 940.0mL of sterile cooled blood agar base No. 2, aseptically add 50.0mL of sterile, lysed defibrinated horse blood and 10.0mL of sterile antibiotic solution. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and cultivation of *Campylobacter* species.

SL Medium

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Glucose	10.0g
KH ₂ PO ₄	6.0g
Arabinose	5.0g
Sucrose	5.0g
Yeast extract	5.0g
Sodium acetate·3H ₂ O	2.5g
Diammonium citrate	2.0g
Tween 80	1.0g
MgSO ₄ ·7H ₂ O	0.58g
MnSO ₄ ·4H ₂ O	0.28g

pH 5.4 ± 0.2 at 25°C

Preparation of Medium: Add agar to 500.0mL of distilled/deionized water. Gently heat and bring to boiling. In a separate flask, add the remaining components, except sodium acetate, to distilled/deionized water and bring volume to 300.0mL. Mix thoroughly. Combine the two solutions. In a separate flask, add sodium acetate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Adjust pH to 5.4 with glacial acetic acid. Add this solution to the agar solution. Mix thoroughly. Adjust pH to 5.4 with additional glacial acetic acid. Bring volume to 1.0L with distilled/deionized water. Do not autoclave. Aseptically distribute into sterile screw-capped tubes or flasks.

Use: For the selective isolation and cultivation of *Lactobacillus* species.

Slad Medium

Composition per liter:

Solution B	900.0mL
Solution A	100.0mL

Solution A:

Composition per 100.0mL:

Raffinose	20.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Ammonium sulfate	7.0mg
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg

Thiamine·HCl	0.4mg
Pyroxidine·HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
p-Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid	2.0μg
Biotin	2.0μg

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°–55°C.

Solution B:

Composition per 900.0mL:

Agar	40.0g

Preparation of Solution B: Add agar to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Preparation of Medium: Aseptically combine 100.0mL of sterile solution A and 900.0mL of sterile solution B. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Slanetz and Bartley Medium

Composition per liter:

Tryptose	20.0g
Agar	10.0g
Yeast extract	5.0g
Na ₂ HPO ₄ 2H ₂ O	4.0g
Glucose	2.0g
NaN ₃	0.4g
Tetrazolium chloride	0.1g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the detection and enumeration of enterococci by the membrane filter method.

SM ID2 Agar

Composition per liter:

Proprietary

Source: This medium is available from bioMérieux

Use: For the selective isolation and differentiation of the genus *Salmonella* spp. in food products. A chromogenic medium for the selective isolation and detection of *Salmonella*. *S. typhi*, and *S. paratyphi*, and most Lactose(+) *Salmonella* present pale pink to mauve colonies. Other organisms are either inhibited, colorless, or pale blue in appearance

Sodium Chloride Broth, 6.5

Composition per liter:

Beef heart, solids from infusion.....	500.0g
NaCl.....	65.0g
Tryptose.....	10.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of enterococci and other salt-tolerant organisms. For the differentiation of microorganisms based on salt tolerance.

Sodium Dodecyl Sulfate

Polymyxin Sucrose Agar

Composition per liter:

NaCl.....	20.0g
Agar.....	15.0g
Sucrose.....	15.0g
Proteose peptone.....	10.0g
Beef extract.....	5.0g
Sodium lauryl sulfate.....	1.0g
Bromthymol Blue.....	0.04g
Cresol Red.....	0.04g
Polymyxin B solution.....	10.0mL

pH 7.6 ± 0.2 at 25°C

Polymyxin B Solution:

Composition per 10.0mL:

Polymyxin B sulfate.....	100,000U

Preparation of Polymyxin B Solution: Add polymyxin B sulfate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except polymyxin B solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Aseptically add sterile polymyxin B solution. Mix thoroughly. Pour immediately into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Vibrio* species from foods.

Sodium Lactate Agar

Composition per liter:

Agar.....	15.0g
Pancreatic digest of casein.....	10.0g
Sodium lactate.....	10.0g
Yeast extract.....	10.0g
K ₂ HPO ₄	0.25g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Adjust pH to 7.0. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and isolation of *Propionibacterium* species from foods. For the isolation and cultivation of propionic acid-producing bacteria from cheese.

Sodium Lactate Agar, Modified

Composition per liter:

Agar.....	15.0g
Yeast extract.....	10.0g
Pancreatic digest of casein.....	5.1g
NaCl.....	1.5g
Papaic digest of soybean meal.....	0.9g
K ₂ HPO ₄	0.75g
Glucose.....	0.75g
Sodium lactate, 60% syrup.....	20.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and isolation of *Propionibacterium* species from foods. For the isolation and cultivation of propionic acid-producing bacteria from cheese.

Soft Agar Gelatin Overlay

Composition per plate:

Base agar.....	15.0mL
Soft agar gelatin overlay.....	2.5mL

pH 7.0 ± 0.2 at 25°C

Base Agar:**Composition per liter:**

Agar	15.0g
Peptone.....	5.0g
NaCl.....	5.0g
Beef extract.....	3.0g
MnSO ₄ ·H ₂ O	0.05g

Preparation of Base Agar: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Soft Agar Gelatin Overlay:**Composition per liter:**

Gelatin.....	15.0g
Agar	8.0g
Peptone.....	5.0g
NaCl.....	5.0g
Beef extract.....	3.0g
MnSO ₄ ·H ₂ O	0.05g

Preparation of Soft Agar Gelatin Overlay: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Aseptically pour cooled, sterile base agar into sterile Petri dishes in 15.0mL volumes. Allow agar to solidify. Inoculate plates with samples. Overlay each plate with 2.5mL of soft agar gelatin overlay.

Use: For the cultivation and differentiation of microorganisms based on proteolytic activity.

Sorbitol Agar**Composition per liter:**

Agar	20.0g
Peptone.....	10.0g
NaCl.....	2.0g
Yeast extract.....	2.0g
Sorbitol solution.....	50.0mL

pH 7.0 ± 0.2 at 25°C

Sorbitol Solution:**Composition per 50.0mL:**

Sorbitol.....	5.0g

Preparation of Sorbitol Solution: Add sorbitol to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except sorbitol solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Adjust pH to 7.0. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to

45°–50°C. Aseptically add sterile sorbitol solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Pseudomonas* species.

**Sorbitol MacConkey Agar
(MacConkey Agar with Sorbitol)**

Composition per liter:

Peptone	20.0g
Agar	15.0g
Sorbitol	10.0g
NaCl.....	5.0g
Bile salts No. 3.....	1.5g
Neutral Red.....	0.03g
Crystal Violet.....	1.0mg

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of pathogenic *Escherichia coli*.

**Sorbitol MacConkey Agar
(MacConkey Agar with Sorbitol)
(BAM M139)**

Composition per liter:

Peptone or gelysate.....	17.0g
Agar	13.5g
Sorbitol	10.0g
NaCl.....	5.0g
Proteose peptone No. 3 or polypeptone.....	3.0g
Bile salts, purified.....	1.5g
Neutral Red.....	0.03g
Crystal Violet.....	1.0mg

pH 7.1 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of pathogenic *Escherichia coli*.

Sorbitol MacConkey Agar with BCIG (SMAC with BCIG)

Composition per liter:

Peptone.....	20.0g
Agar	15.0g
Sorbitol.....	10.0g
NaCl.....	5.0g
Protease peptone.....	3.0g
Bile salts mixture	1.5g
5-Bromo-4-chloro-3-indolyl- β -D-glucuronide sodium salt.....	0.1g
Neutral Red.....	0.03g
pH 7.1 $\pm$ 0.2 at 25°C	

Source: This medium is available from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: A selective and differential medium for the detection of *Escherichia coli* O157 incorporating the chromogen 5-bromo-4-chloro-3-indolyl- β -D-glucuronide (BCIG). The medium combines two different screening mechanisms for the detection of *E. coli* O157, the failure to ferment sorbitol and the absence of β -glucuronidase activity. The non-sorbitol-fermenting and β -glucuronidase-negative *E. coli* O157 will appear as straw colored colonies. Organisms with β -glucuronidase activity will cleave the substrate, leading to a distinct blue-green coloration of the colonies. The intestinal tract of ruminants is the prime reservoir of *E. coli* O157 and other enterohaemorrhagic *E. coli* (EHEC) strains, therefore meats derived from cattle, sheep, goat, and deer can be expected to be contaminated. Foods implicated in human illness related to *E. coli* O157 include meats, dairy products, vegetables, salads, apple juice, and water.

Sour Dough Medium

Composition per liter:

Maltose.....	20.0g
Pancreatic digest of casein.....	6.0g
Yeast extract.....	3.0g
Tween 80.....	0.3g
Fresh yeast extract solution.....	15.0mL
pH 5.6 $\pm$ 0.2 at 25°C	

Fresh Yeast Extract Solution:

Composition per 100.0mL:

Baker's yeast, live, pressed, starch-free.....	25.0g

Preparation of Fresh Yeast Extract Solution: Add the live Baker's yeast to 100.0mL of distilled/deionized water. Autoclave for 90 min at 15 psi pres-

sure–121°C. Allow to stand. Remove supernatant solution. Adjust pH to 6.6–6.8.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.6 with 20% lactic acid or 6N HCl. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus sanfrancisco*.

Spirit Blue Agar

Composition per liter:

Agar	20.0g
Pancreatic digest of casein.....	10.0g
Yeast extract.....	5.0g
Spirit Blue.....	0.15g
Lipoidal emulsion	30.0mL
pH 6.8 $\pm$ 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Lipoidal Emulsion:

Composition per 500.0mL:

Tween 80.....	1.0mL
Cottonseed oil or olive oil.....	100.0mL

Preparation of Lipoidal Emulsion: Add Tween 80 to 400.0mL of warm distilled/deionized water. Mix thoroughly. Add 100.0mL of cottonseed or olive oil. Emulsify in a blender. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Preparation of Medium: Add components, except lipoidal emulsion, to distilled/deionized water and bring volume to 970.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 30.0mL of sterile lipoidal emulsion. Mix thoroughly. Pour into sterile Petri dishes while shaking flask to keep emulsion dispersed.

Use: For the detection, enumeration, and study of lipolytic microorganisms.

Spore Strip Broth

Composition per liter:

Spore strip broth	9.0g

Preparation of Medium: Add 9.0g of spore strip broth powder (a mixture of glucose, buffer salts, growth factors, and Bromthymol Blue) to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the recovery of spores of *Bacillus stearothermophilus* on spore strips used to determine the sterilization efficiency of autoclaves.

Sporulation Broth (BAM M140)

Composition per liter:

Polypeptone	15.0g
Na ₂ HPO ₄	11.0g
Starch, soluble	3.0g
Yeast extract	3.0g
Sodium thioglycolate	1.0g
MgSO ₄ , anhydrous	0.1g

pH 7.8 ± 0.1 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 15.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and observation of sporulation of *Clostridium perfringens*.

Spray's Fermentation Medium (BAM M141)

Composition per 1100.0mL:

Neopeptone	10.0g
Pancreatic digest of casein	10.0g
Agar	2.0g
Sodium thioglycolate	0.025g
Carbohydrate solution	110.0mL

pH 7.4 ± 0.1 at 25°C

Carbohydrate Solution:

Composition per 200.0mL:

Carbohydrate	20.0g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 200.0mL. Glucose or glycerol may be used. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except agar and carbohydrate solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Adjust pH to 7.4. Add agar. Gently heat and bring to boiling. Distribute into tubes in 9.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Immediately prior to use, heat tubes in a boiling water bath for 10 min. Cool to 45°C. Aseptically add 1.0mL of sterile carbohydrate solution. Mix thoroughly.

Use: For the cultivation and differentiation of *Clostridium perfringens* based on carbohydrate fermentation patterns.

SPS Agar

(Sulfite Polymyxin Sulfadiazine Agar)

Composition per liter:

Pancreatic digest of casein	15.0g
Agar	13.9g
Yeast extract	10.0g
Ferric citrate	0.5g
Na ₂ SO ₃	0.5g
Polymyxin sulfate	0.01g
Sulfadiazine	0.12g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and detection of *Clostridium perfringens* and *Clostridium botulinum* in foods and other materials.

SS Deoxycholate Agar (*Salmonella-Shigella* Deoxycholate Agar) (SSDC)

Composition per liter:

Agar	13.5g
Lactose	10.0g
Sodium deoxycholate	10.0g
Bile salts	8.5g
Na ₂ S ₂ O ₃	8.5g
Sodium citrate	8.5g
Beef extract	5.0g
Pancreatic digest of casein	2.5g
Peptic digest of animal tissue	2.5g
CaCl ₂ ·2H ₂ O	1.0g
Ferric citrate	1.0g
Neutral Red	0.025g
Brilliant Green	0.33mg

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes in 20.0mL volumes. Allow the surface of the plates to dry before inoculation.

Use: For the isolation and cultivation of *Yersinia enterocolitica* from foods.

SSM Agar**Composition per liter:**

Solution A	850.0mL
Solution C	100.0mL
Solution B	50.0mL

Solution A:**Composition per 850.0mL:**

Agar, noble	15.0g
Na ₂ HPO ₄	6.0g
KH ₂ PO ₄	3.0g
MgSO ₄ ·7H ₂ O	0.2g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 850.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution B:**Composition per 50.0mL:**

Glucose	5.0g

Preparation of Solution B: Add glucose to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Solution C:**Composition per 100.0mL:**

Casein hydrolysate	20.0g

Preparation of Solution C: Add casein hydrolysate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter through Whatman #1 paper. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Preparation of Medium: Aseptically combine 850.0mL of sterile solution A, 50.0mL of sterile solution B, and 100.0mL of sterile solution C. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and maintenance of a variety of bacteria.

**ST Holding Medium
(m-ST Holding Medium)**

Composition per liter:

KH ₂ PO ₄	3.0g
Tris(hydroxymethyl)aminomethane buffer	3.0g
Sulfanilamide	1.5g
NaH ₂ PO ₄ ·H ₂ O	0.1g
Ethanol (95% solution)	10.0mL
pH 8.6 ± 0.2 at 25°C	

Preparation of Medium: Dissolve the sulfanilamide in the ethanol. Add all components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–

121°C. Distribute in 1.8mL volumes to sterile Petri dishes with tight-fitting lids and an absorbent filter.

Use: For the cultivation and enumeration of coliform bacteria by the delayed-incubation total coliform procedure. For use as a holding or transport medium to keep coliform bacteria viable between sampling and laboratory culture.

STAA Agar Base**Composition per liter:**

Peptone	20.0g
Agar	13.0g
Glycerol	7.5g
Yeast extract	2.0g
K ₂ HPO ₄	1.0g
MgSO ₄ ·7H ₂ O	1.0g
STAA selective supplement	10.0mL
pH 7.0 ± 0.1 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

STAA Selective Supplement:**Composition per 10.0mL:**

Streptomycin sulfate	0.5g
Cycloheximide	0.05g
Thallous acetate	0.05g

Preparation of STAA Selective Supplement:

Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except glycerol and STAA selective supplement, to distilled/deionized water and bring volume to 985.0mL. Mix thoroughly. Gently heat and bring to boiling. Add glycerol. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile STAA selective supplement. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation of *Brochothrix thermosphacta* from meat products.

**Standard Methods Agar
(Tryptone Glucose Yeast Agar)
(Plate Count Agar)**

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	5.0g
Yeast extract	2.5g
Glucose	1.0g
pH 7.0 ± 0.1 at 25°C	

Source: Available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration by microbial plate counts of microorganisms isolated from milk and dairy products, foods, water, and other specimens.

Standard Methods Agar with Lecithin and Polysorbate 80

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	5.0g
Polysorbate 80.....	5.0g
Yeast extract.....	2.5g
Glucose	1.0g
Lecithin	0.7g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For determination of the sterility of surfaces.

Standard Methods Broth (m-Standard Methods Broth) (Tryptone Glucose Yeast Broth) (m-Plate Count Broth)

Composition per liter:

Pancreatic digest of casein.....	10.0g
Yeast extract.....	5.0g
Glucose	2.0g

pH 7.0 ± 0.2 at 25°C

Source: Available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For enumeration of the total number of microorganisms by the membrane filter method.

Standard Methods Caseinate Agar

Composition per liter:

Agar	15.0g
Sodium caseinate	10.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	2.5g
Glucose	1.0g
Trisodium citrate solution.....	1.0L
CaCl ₂ ·2H ₂ O (1M solution).....	20.0mL

Trisodium Citrate Solution:

Composition per liter:

Trisodium citrate·2H ₂ O.....	4.41g

Preparation of Trisodium Citrate Solution:

Add trisodium citrate·2H₂O to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add yeast extract, pancreatic digest of casein, glucose, and agar to 500.0mL of the trisodium citrate solution. Mix thoroughly. Gently heat until boiling. Add the remaining components. Bring volume to 1.0L with trisodium citrate solution. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the cultivation of proteolytic bacteria from foods.

Standard II Nutrient Agar

Composition per liter:

Agar	13.0g
Tryptose	7.0g
NaCl.....	5.0g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of nonfastidious microorganisms. For the maintenance of cultures of a wide variety of nonfastidious bacteria. May also be used as a base for blood and other enrichments for the cultivation of fastidious microorganisms. May be used to determine indole production.

Staphylococcus Agar No. 110 (BAM M142)

Composition per liter:

NaCl.....	75.0g
Gelatin.....	30.0g

Agar	15.0g
D-Mannitol	10.0g
Pancreatic digest of casein	10.0g
K ₂ HPO ₄	5.0g
Yeast extract	2.5g
Lactose	2.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Swirl flask while pouring plates to disperse precipitate.

Use: For the isolation and enumeration of staphylococci.

Staphylococcus Broth (*m-Staphylococcus Broth*)

Composition per liter:

NaCl	75.0g
Mannitol	10.0g
Pancreatic digest of casein	10.0g
K ₂ HPO ₄	5.0g
Yeast extract	2.5g
Lactose	2.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and enumeration of pathogenic and enterotoxigenic staphylococci by the membrane filter method.

Staphylococcus Medium

Composition per liter:

Agar	15.0g
Peptone	6.0g
Pancreatic digest of casein	4.0g
Yeast extract	3.0g
Beef extract	1.5g
Glucose	1.0g
pH 6.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Staphylococcus aureus*. For the enumeration of pathogenic and enterotoxigenic staphylococci by the membrane filter method.

Staphylococcus Medium No. 110

Composition per liter:

NaCl	75.0g
Gelatin	30.0g
Agar	15.0g
Mannitol	10.0g
Pancreatic digest of casein	10.0g
K ₂ HPO ₄	5.0g
Yeast extract	2.5g
Lactose	2.0g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective isolation, cultivation, and maintenance of *Staphylococcus* species.

Staphylococcus/Streptococcus Selective Medium

Composition per 1060.0mL:

Columbia blood agar base	1.0L
Horse blood, defibrinated	50.0mL
Antibiotic inhibitor	10.0mL
pH 7.3 ± 0.2 at 25°C	

Columbia Blood Agar Base:

Composition per liter:

Special peptone	23.0g
Agar	10.0g
NaCl	5.0g
Starch	1.0g

Source: Columbia blood agar base is available as a premixed powder from Oxoid.

Preparation of Columbia Blood Agar Base:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Antibiotic Inhibitor:**Composition** per 10.0mL:

Nalidixic acid.....	0.015g
Colistin sulfate.....	0.01g
Ethanol (95% solution).....	10.0mL

Preparation of Antibiotic Inhibitor: Add components to 10.0mL ethanol. Mix thoroughly. Filter sterilize.

Preparation of Medium: To 1.0L of cooled sterile Columbia blood agar base, aseptically add sterile horse blood and sterile antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of *Staphylococcus aureus* and streptococci from Starch Agar

Composition per liter:

Agar.....	12.0g
Soluble starch.....	10.0g
Beef extract.....	3.0g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and differentiation of a variety of microorganisms based on amylase production. After incubation, starch hydrolysis is determined by the addition of Gram's or Lugol's iodine solution. Organisms that produce amylase appear as colonies surrounded by a clear zone.

Starch Agar**Composition** per liter:

Agar.....	12.0g
Soluble starch.....	10.0g
Beef extract.....	3.0g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and differentiation of a variety of microorganisms based on amylase production. After incubation, starch hydrolysis is determined by the addition of Gram's or Lugol's iodine solution. Or-

ganisms that produce amylase appear as colonies surrounded by a clear zone.

Starch Agar**(BAM M143)****Composition** per liter:

Agar.....	15.0g
Soluble starch.....	10.0g
Pancreatic digest of gelatin.....	5.0g
Beef extract.....	3.0g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a wide variety of bacteria.

Starch Hydrolysis Agar**Composition** per liter:

Beef heart, infusion from.....	500.0g
Soluble starch.....	20.0g
Agar.....	15.0g
Tryptose.....	10.0g
NaCl.....	5.0g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and differentiation of a variety of microorganisms based on amylase production. After incubation, starch hydrolysis is determined by the addition of Gram's or Lugol's iodine solution. Organisms that produce amylase appear as colonies surrounded by a clear zone.

Sterility Test Broth**(USP Alternative****Thioglycolate Medium)****Composition** per liter:

Pancreatic digest of casein.....	15.0g
Glucose.....	5.0g
Yeast extract.....	5.0g
NaCl.....	2.5g

L-Cystine	0.5g
Sodium thioglycolate	0.5g
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 25°C . If not used immediately, prior to inoculation heat tubes in a boiling water bath for 5–10 min. Cool to 25°C .

Use: As an alternate medium, instead of fluid thioglycolate broth, for testing the sterility of a variety of specimens.

Stock Culture Agar

Composition per liter:

Beef heart infusion.....	500.0g
Gelatin.....	10.0g
Proteose peptone.....	10.0g
Agar	7.5g
Casein.....	5.0g
Na_2HPO_4	4.0g
Sodium citrate	3.0g
Glucose	0.5g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to cold distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the maintenance of pathogenic and non-pathogenic bacteria, especially streptococci.

Stock Culture Agar with L-Asparagine

Composition per liter:

Beef heart infusion.....	500.0g
Gelatin.....	10.0g
Proteose peptone.....	10.0g
Agar	7.5g
Casein.....	5.0g
Na_2HPO_4	4.0g
Sodium citrate	3.0g
L-Asparagine	1.0g
Glucose	0.5g
pH 7.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to cold distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure– 121°C . Pour into sterile Petri dishes or leave in tubes.

Use: For the maintenance of pathogenic and non-pathogenic bacteria, especially streptococci.

Stonebrink's Medium

Composition per 1600.0mL:

Homogenized whole egg	2.0L
Mineral salts solution.....	1.0L
Malachite green solution.....	40.0mL

Mineral Salts Solution:

Na-pyruvate	12.5g
KH_2PO_4	7.0g
$\text{Na}_2\text{HPO}_4 \cdot 7\text{H}_2\text{O}$	4.0g

Preparation of Mineral Salts Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 50°C .

Malachite Green Solution:

Composition per 100.0mL:

Malachite green	2.0g

Preparation of Malachite Green Solution:

Add malachite to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure– 121°C . Cool to 50°C .

Homogenized Whole Egg:

Composition per 2.0L:

Whole eggs	36-48

Preparation of Homogenized Whole Egg:

Use fresh eggs, less than 1 week old. Scrub the shells with soap. Let stand in a soap solution for 30 min. Rinse in running water. Soak eggs in 70% ethanol for 15 min. Break the eggs into a sterile container. Homogenize by shaking. Filter through four layers of sterile cheesecloth into a sterile graduated cylinder. Measure out 2.0L.

Preparation of Medium: Aseptically add 40.0mL sterile malachite to 1.0L of sterile mineral salts solution. Mix thoroughly. Aseptically add 2.0L of homogenized whole egg. Mix thoroughly. Distribute into sterile screw-capped tubes. Place tubes in a slanted position. Inspissate at 85°C (moist heat) for 45 min.

Use: For the cultivation of *Mycobacterium* species, especially bovine strains. For the primary isolation of *Mycobacterium bovis*.

Strep ID Quad Plate**Composition per liter:**

Quadrant I	5.0mL
Quadrant II	5.0mL
Quadrant III	5.0mL
Quadrant IV	5.0mL

Source: Available as a prepared medium from BD Diagnostic Systems.

Quadrant I:**Composition per 5.0mL:**

Bacitracin	0.5mg
TSA II agar	5.0mL

Quadrant II:**Composition per 5.0mL:**

TSA II agar	5.0mL
Sheep blood, defibrinated	0.25mL

Quadrant III:**Composition per 5.0mL:**

Bile esculin agar	5.0mL

Quadrant IV:**Composition per 5.0mL:**

Blood agar base with 6.5% NaCl	5.0mL

Use: For the differentiation and presumptive identification of streptococci. The Strep (*Streptococcus*) ID (Identification) Quad Plate is a four-sectored plate. Each plate sector contains a different medium.

Streptococcal Growth Medium**Composition per liter:**

Beef heart, solids from infusion	500.0g
Tryptose	10.0g
NaCl	5.0g
Glucose	1.0g
Bromcresol Purple solution	1.0mL

pH 7.4 ± 0.2 at 25°C

Bromcresol Purple Solution:**Composition per 10.0mL:**

Bromcresol Purple	0.16g
Ethanol (95% solution)	10.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to 10.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Streptococcus* species and other Gram-positive cocci. Growth in this medium turns the indicator yellow and the solution turbid.

Streptococcus Agar**Composition per liter:**

Glucose	20.0g
Pancreatic digest of casein	20.0g
Agar	15.0g
K ₂ HPO ₄	2.0g
MgSO ₄ ·7H ₂ O	0.1g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Streptococcus* species.

Streptococcus Medium**Composition per liter:**

Agar	15.0g
Glucose	4.0g
K ₂ HPO ₄	3.8g
Pancreatic digest of casein	2.5g
Yeast extract	2.5g

pH 7.6 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Enterococcus faecalis*.

Streptococcus Selective Medium**Composition per liter:**

Special peptone	23.0g
Agar	10.0g
NaCl	5.0g
Starch	1.0g
Horse blood, defibrinated	50.0mL
Antibiotic inhibitor	10.0mL

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Antibiotic Inhibitor:**Composition per 10.0mL:**

Colistin sulfate	10.0mg
Oxolinic acid	5.0mg

Preparation of Antibiotic Inhibitor: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse blood and antibiotic inhibitor, to distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile horse blood and sterile antibiotic inhibitor. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of streptococci from clinical specimens or foodstuffs.

***Streptococcus thermophilus* Agar (ST Agar)**

Composition per liter:

Agar	15.0g
Sucrose	10.0g
Pancreatic digest of casein	10.0g
Yeast extract	5.0g
K ₂ HPO ₄	2.0g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Streptococcus thermophilus* from dairy products.

Succinate Mineral Medium

Composition per 1001.0mL:

MnSO ₄ ·7H ₂ O	62.0g
K ₂ HPO ₄ ·3H ₂ O	1.63g
NH ₄ Cl	1.02g
KH ₂ PO ₄	0.39g
Succinate	10.0mM
Trace elements solution	1.0mL

pH 6.8 ± 0.2 at 25°C

Trace Elements Solution:

Composition per 100.0mL:

Solution A	50.0mL
Solution B	50.0mL

Preparation of Trace Elements Solution:

Aseptically combine 50.0mL of sterile solution A with 50.0mL of sterile solution B.

Solution A:

Composition per liter:

Disodium EDTA	0.25g
CaCl ₂ ·2H ₂ O	0.14g
FeCl ₂ ·4H ₂ O	0.14g

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Solution B:

Composition per liter:

Disodium EDTA	0.25g
H ₃ BO ₃	30.0mg
CoCl ₂ ·2H ₂ O	28.0mg
ZnCl ₂	5.0mg
MnCl ₂ ·4H ₂ O	4.0mg
NaMoO ₄ ·2H ₂ O	3.0mg
NiCl ₂ ·6H ₂ O	1.65mg
CuCl ₂ ·2H ₂ O	0.7mg

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C.

Preparation of Medium: Add components, except trace elements solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 1.0mL of sterile trace elements solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of bacteria that can utilize succinate as a carbon source.

Sucrose Agar

Composition per liter:

Sucrose	50.0g
Agar	20.0g
Peptone	10.0g
NaCl	5.0g
Yeast extract	5.0g
CaCO ₃	3.0g
Phenylethyl alcohol	3.0g
MgSO ₄ ·7H ₂ O	0.5g
MnSO ₄ ·4H ₂ O	0.5g
Tween 80	0.1g
Bromcresol Green	20.0mg
Cycloheximide solution	10.0mL

pH 6.2 ± 0.2 at 25°C

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.01g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add agar to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Add remaining components, except cycloheximide solution and phenylethyl alcohol, and bring volume to

990.0mL with distilled/deionized water. Adjust pH to 6.2. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile cycloheximide solution and 3.0g of phenylethyl alcohol. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Lactobacillus* species from brewery isolates.

Sucrose Teepol Tellurite Agar (STT Agar)

Composition per liter:

Agar	20.0g
Beef extract	1.0g
Peptone	1.0g
Sucrose	1.0g
NaCl	0.5g
Bromthymol Blue (0.2% solution)	2.5mL
Tellurite solution	2.5mL
Sodium lauryl sulfate (Teepol, 0.1% solution)	0.2mL
pH 8.0 ± 0.2 at 25°C	

Tellurite Solution:

Composition per 100.0mL:

K ₂ TeO ₃	0.05g

Preparation of Tellurite Solution: Add the K₂TeO₃ to distilled/deionized water and bring the volume to 100.0mL. Mix thoroughly. Filter sterilize. Use freshly prepared solution.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Pour into sterile Petri dishes.

Use: For the selective isolation, cultivation, and differentiation of *Vibrio* species based on their ability to ferment sucrose. *Vibrio cholerae* appears as flat yellow colonies. *Vibrio parahaemolyticus* appears as levated green-yellow mucoid colonies.

Sucrose Yeast Extract Medium

Composition per liter:

Sucrose	20.0g
Yeast extract	4.0g
K ₂ HPO ₄	2.5g
MgSO ₄ ·7H ₂ O	1.0g
Trace elements solution	4.0mL
pH 7.0 ± 0.2 at 25°C	

Trace Elements Solution:

Composition per liter:

ZnSO ₄ ·7H ₂ O	18.0g
FeSO ₄ ·7H ₂ O	9.0g

MnSO ₄ ·4H ₂ O	3.0g
CoCl ₂ ·6H ₂ O	0.9g
CuSO ₄ ·5H ₂ O	0.8g
H ₂ SO ₄ (Concentrated)	5.0mL

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of bacteria that can utilize sucrose as carbon source.

Superbroth

Composition per liter:

Pancreatic digest of casein	32.0g
Yeast extract	20.0g
NaCl	5.0g
NaOH (1N solution)	5.0mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli*.

Syncase Broth

Composition per liter:

Casamino acids	20.0g
K ₂ HPO ₄	8.71g
Yeast extract	6.0g
NaCl	2.5g

pH 8.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.5. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of heat-labile, toxin-producing *Escherichia coli* from foods.

T₁N₀ Broth (Tryptone Broth) (BAM M161)

Composition per liter:

Pancreatic digest of casein	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₁ Agar
(Tryptone Salt Agar)
(BAM M144)/(BAM M163)

Composition per liter:

Agar	20.0g
NaCl.....	10.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₂ Agar
(Tryptone Salt Agar)
(BAM M163)

Composition per liter:

Agar	20.0g
NaCl.....	20.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₁ Broth
(Tryptone Salt Broth)
(BAM M161)

Composition per liter:

NaCl.....	10.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling.

Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₃ Broth
(Tryptone Salt Broth)
(BAM M161)

Composition per liter:

NaCl.....	30.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₆ Broth
(Tryptone Salt Broth)
(BAM M161)

Composition per liter:

NaCl.....	60.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₈ Broth
(Tryptone Salt Broth)
(BAM M161)

Composition per liter:

NaCl.....	80.0g
Pancreatic digest of casein.....	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T₁N₁₀ Broth (Tryptone Salt Broth) (BAM M161)

Composition per liter:

NaCl	100.0g
Pancreatic digest of casein	10.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Vibrio cholerae* and other *Vibrio* species.

T7 Agar Base (m-T7 Agar Base)

Composition per liter:

Lactose	20.0g
Agar	15.0g
Polyoxyethylene ether W-1	5.0g
Yeast extract	3.0g
Pancreatic digest of casein	2.5g
Peptic digest of animal tissue	2.5g
Sodium heptadecyl sulfate	0.1g
Bromthymol Blue	0.1g
Bromocresol Purple	0.1g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45–50°C. The medium may be made more selective by adding 1.0mg of penicillin G per liter. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective recovery and differential identification of injured coliform microorganisms from chlorinated water by the membrane filter method. For rapid estimation of the bacteriological quality of water using the membrane filter method.

Tap Water Agar

Composition per liter:

Agar	15.0g
Tap water	1.0L

Preparation of Medium: Add agar to 1.0L of tap water. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and differentiation of fungi and aerobic actinomycetes based on filament and aerial hyphae morphology.

Tarshis Blood Agar

Composition per 1050.0mL:

Beef heart infusion	500.0g
Agar	15.0g
Meat peptone	10.0g
NaCl	5.0g
Penicillin G, sterile	100,000U
Sheep blood, sterile	300.0mL
Glycerol	10.0mL
pH 6.6 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood and penicillin G, to distilled/deionized water and bring volume to 750.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45–50°C. Aseptically add sterile sheep blood and sterile penicillin G. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Mycobacterium tuberculosis*.

TB Nitrate Reduction Broth

Composition per 100.0mL:

Na ₂ HPO ₄ ·12H ₂ O	0.485g
KH ₂ PO ₄	0.117g
NaNO ₃	0.085g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of *Mycobacterium* species based on nitrate reduction. After growth of cells in appropriate medium, nitrate reduction is determined by making a suspension of cells in TB nitrate reduction broth and adding hydrochloric acid, sulfanilamide, and *N*-naphthylenediamine. Nitrate reduction turns the medium pink. *Mycobacterium tuberculosis* reduces nitrate and turns the medium deep pink within 1 min. *Mycobacterium bovis* does not reduce nitrate and does not change the medium.

TBX Agar

(Tryptone Bile X-glucuronide Agar) (TBX Chromogen Medium)

Composition per liter:

Peptone	20.0g

Agar	15.0g
Bile salts.....	1.5g
X-β-D-glucuronide.....	0.075g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available from Fluka, Sigma-Aldrich and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the detection and enumeration of *E. coli* in foodstuffs, animal food and water without further confirmation. *E. coli* colonies are colored blue-green. Unlike MUG, where the fluorophore leaches out of the cell into the surrounding agar, the released chromophore in TBX Medium is insoluble and accumulates within the cell. This ensures that colored target colonies are easy to identify. Most *E. coli* strains can be differentiated from other coliforms by the presence of the enzyme glucuronidase. The chromogen in TBX Medium is 5-bromo-4-chloro-3-indolyl-β-D-glucuronide (X-glucuronide), and is targeted by this enzyme. *E. coli* cells are able to absorb this complex intact and intracellular glucuronidase splits the bond between the chromophore and the glucuronide. The released chromophore is colored and builds up within the cells, causing *E. coli* colonies to be colored blue/green.

TBYA Agar (Tryptone Beef Yeast Extract Acetate Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	2.0g
Beef extract.....	0.5g
Yeast extract.....	0.5g
Sodium acetate.....	0.2g
pH 7.2-7.4 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2-7.4. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Leuconostoc* species.

TCBS Agar (Thiosulfate Citrate Bile Salt Sucrose Agar) (BAM M147)

Composition per liter:

Sucrose.....	20.0g
Agar	14.0g
NaCl.....	10.0g
Sodium citrate.....	10.0g
Na ₂ S ₂ O ₃	10.0g
Yeast extract.....	5.0g
Pancreatic digest of casein.....	5.0g
Peptic digest of animal tissue	5.0g
Oxgall	5.0g
Sodium cholate	3.0g
Ferric citrate.....	1.0g
Thymol Blue	0.04g
Bromthymol Blue	0.04g
pH 8.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of *Vibrio cholerae* and *Vibrio parahaemolyticus* from a variety of foods.

TCH Medium (Thiophene 2 Carboxylic Acid Hydrazide Medium)

Composition per 1105.0mL:

Thiophene-2-carboxylic acid hydrazide	1.1mg
Middlebrook 7H10 agar base.....	1.0L
OADC enrichment.....	100.0mL
Glycerol	5.0mL
pH 6.6 ± 0.2 at 25°C	

Middlebrook 7H10 Agar Base:

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄	1.5g
KH ₂ PO ₄	1.5g
(NH ₄) ₂ SO ₄	0.5g
L-Glutamic acid.....	0.5g
Sodium citrate.....	0.4g
Ferric ammonium citrate.....	0.04g
MgSO ₄ ·7H ₂ O	0.025g
ZnSO ₄ ·7H ₂ O	1.0mg
CuSO ₄ ·5H ₂ O	1.0mg
Pyridoxine.....	1.0mg
Biotin	0.5mg

CaCl ₂ ·2H ₂ O.....	0.5mg
Malachite Green.....	0.25mg

Preparation of Middlebrook 7H10 Agar

Base: Add glycerol to 900.0mL of distilled/deionized water and add remaining components. Mix thoroughly. Gently heat and bring to boiling.

Middlebrook OADC Enrichment:

Composition per 100.0mL:

Bovine albumin fraction V	5.0g
Glucose	2.0g
NaCl.....	0.85g
Oleic acid.....	0.05g
Catalase.....	4.0mg

Source: This enrichment is available as a prepared enrichment from BD Diagnostic Systems.

Preparation of Middlebrook OADC Enrichment:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation for Medium: Combine components. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of *Mycobacterium* species based on sensitivity to TCH. *Mycobacterium bovis* is inhibited by TCH. *Mycobacterium tuberculosis* and other mycobacteria are generally resistant to low concentrations of TCH. This distinguishes *Mycobacterium bovis* from other nonchromogenic, slow-growing mycobacteria.

Teepol Broth, Enriched (m-Teepol Broth, Enriched)

Composition per liter:

Peptone.....	40.0g
Lactose	30.0g
Yeast extract.....	6.0g
Phenol Red.....	0.2g
Sodium lauryl sulfate (Teepol, 0.1% solution)	4.0mL
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enumeration of coliform organisms and *Escherichia coli* by the membrane filter method.

Tellurite Glycine Agar

Composition per liter:

Agar	17.5g

Pancreatic digest of casein.....	10.0g
Glycine.....	10.0g
Yeast extract.....	6.5g
D-Mannitol	5.0g
K ₂ HPO ₄	5.0g
LiCl.....	5.0g
Enzymatic hydrolysate of soybean meal	3.5g
Chapman tellurite solution.....	10.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Chapman Tellurite Solution:

Composition per 100.0mL:

K ₂ TeO ₃	1.0g

Preparation of Chapman Tellurite Solution:

Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components, except Chapman tellurite solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 10.0mL of sterile Chapman tellurite solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Allow the surface of the plates to dry before inoculating.

Use: For the isolation and cultivation of coagulase-positive staphylococci.

Tellurite Glycine Agar

Composition per liter:

Agar	16.0g
Pancreatic digest of casein.....	10.0g
Glycine.....	10.0g
Yeast extract.....	5.0g
D-Mannitol	5.0g
K ₂ HPO ₄	5.0g
LiCl.....	5.0g
Chapman tellurite solution.....	20.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Chapman Tellurite Solution:

Composition per 100.0mL:

K ₂ TeO ₃	1.0g

Preparation of Chapman Tellurite Solution:

Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components, except Chapman tellurite solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 20.0mL of sterile Chapman tellurite solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes. Allow the surface of the plates to dry before inoculating.

Use: For the quantitative detection of coagulase-positive staphylococci from foods and other sources.

Tergitol 7 Agar

Composition per liter:

Lactose	20.0g
Agar	13.0g
Peptone.....	10.0g
Yeast extract.....	6.0g
Meat extract	5.0g
Tergitol-7.....	0.1g
Bromthymol Blue	0.05g
TTC solution	5.0mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

TTC Solution:

Composition per 100.0mL:

Triphenyltetrazolium chloride	0.05g

Preparation of TTC Solution: Add triphenyltetrazolium chloride to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 995.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 5.0mL of sterile TTC solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the detection and enumeration of coliforms. Lactose-fermenting bacteria appear as yellow colonies. Lactose-nonfermenting bacteria appear as blue colonies.

Tergitol 7 Agar

Composition per liter:

Agar	15.0g
Lactose	10.0g
Yeast extract.....	3.0g
Pancreatic digest of casein.....	2.5g
Peptic digest of animal tissue.....	2.5g
Tergitol 7	0.1g

Bromthymol Blue	25.0mg
TTC solution.....	3.0mL
pH 6.9 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

TTC Solution:

Composition per 100.0mL:

Triphenyl tetrazolium chloride	1.0g

Preparation of TTC Solution: Add triphenyltetrazolium chloride to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 997.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 3.0mL of sterile TTC solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation and differentiation of coliform bacteria based on lactose fermentation. Lactose-fermenting bacteria appear as yellow colonies. Lactose-nonfermenting bacteria appear as blue colonies.

Tergitol 7 Broth

Composition per liter:

Lactose	10.0g
Yeast extract.....	3.0g
Pancreatic digest of casein.....	2.5g
Peptic digest of animal tissue	2.5g
Tergitol 7.....	0.1g
Bromthymol Blue	25.0mg
TTC solution.....	3.0mL

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

TTC Solution:

Composition per 100.0mL:

Triphenyl tetrazolium chloride	1.0g

Preparation of TTC Solution: Add triphenyltetrazolium chloride to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 997.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 25°C. Aseptically add 3.0mL of sterile TTC solution. Mix thoroughly.

Use: For the isolation and cultivation of coliforms, *Salmonella*, and other enteric bacteria.

Tetrathionate Broth

Composition per liter:

Na ₂ S ₂ O ₃	40.7g
CaCO ₃	25.0g
NaCl	4.5g
Peptone	4.5g
Yeast extract	1.8g
Beef extract	0.9g
Iodine solution	20.0mL

Iodine Solution:

Composition per 20.0mL:

Iodine	6.0g
KI	5.0g

Preparation of Iodine Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 20.0mL of iodine solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the selective isolation and enrichment of *Salmonella typhi* and other salmonellae.

Tetrathionate Broth (BAM M145)

Composition per 1030.0mL:

Tetrathionate broth base	1.0L
Iodine-potassium iodide solution	20.0mL
Brilliant Green solution	10.0mL
pH 8.4 ± 0.2 at 25°C	

Tetrathionate Broth Base:

Composition per liter:

Na ₂ S ₂ O ₃ ·5H ₂ O	30.0g
CaCO ₃	10.0g
Polypeptone	5.0g
Bile salts	1.0g

Preparation of Tetrathionate Broth Base:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. A slight precipitate will remain. Do not autoclave. Cool to 25°C. Store at 4°C.

Iodine-Potassium Iodide Solution:

Composition per 20.0mL:

Iodine, resublimed	6.0g
KI	5.0g

Preparation of Iodine-Potassium Iodide Solution: Add KI to 5.0mL of sterile distilled/deionized water. Mix thoroughly. Add iodine. Mix

thoroughly. Bring volume to 20.0mL with sterile distilled/deionized water.

Brilliant Green Solution:

Composition per 100.0mL:

Brilliant Green	0.1g

Preparation of Brilliant Green Solution: Add Brilliant Green to sterile distilled/deionized water. Mix thoroughly.

Preparation of Medium: Combine 1.0L of tetrathionate broth base, 20.0mL of iodine-potassium iodide solution, and 10.0mL of Brilliant Green solution. Mix thoroughly. Aseptically distribute into tubes in 10.0mL volumes. Do not heat medium after it has been mixed.

Use: For the selective isolation and cultivation of *Salmonella* species from foods.

Tetrathionate Broth (TT Broth)

Composition per liter:

Na ₂ S ₂ O ₃	30.0g
CaCO ₃	10.0g
Proteose peptone	5.0g
Bile salts	1.0g
Iodine solution	20.0mL
pH 8.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Iodine Solution:

Composition per 20.0mL:

Iodine	6.0g
KI	5.0g

Preparation of Iodine Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 20.0mL of iodine solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the selective isolation and enrichment of *Salmonella typhi* and other salmonellae.

Tetrathionate Broth (m-Tetrathionate Broth) (m-TT Broth)

Composition per liter:

Na ₂ S ₂ O ₃	30.0g

CaCO ₃	10.0g
Pancreatic digest of casein	2.5g
Peptic digest of animal tissue	2.5g
Iodine-iodide solution	20.0mL
pH 8.0 ± 0.2 at 25°C	

Iodine-Iodide Solution:**Composition per 20.0mL:**

Iodine	6.0g
KI	5.0g

Preparation of Iodine-Iodide Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine-iodide solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 20.0mL of iodine-iodide solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the selective isolation in the membrane filter method of *Salmonella* species from foods.

Tetrathionate Broth (m-Tetrathionate Broth)

Composition per liter:

Na ₂ S ₂ O ₃	30.0g
Proteose peptone	5.0g
Bile salts	1.0g
Iodine solution	20.0mL
pH 8.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Iodine Solution:**Composition per 20.0mL:**

Iodine	6.0g
KI	5.0g

Preparation of Iodine Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 20.0mL of iodine solution. Mix thoroughly. Use medium the same day it is prepared.

Use: For the enrichment of *Salmonella* species in the membrane filter method prior to placing the filter on selective media such as Brilliant Green broth.

Tetrathionate Broth, Hajna (TT Broth, Hajna)

Composition per liter:

Na ₂ S ₂ O ₃	38.0g
CaCO ₃	25.0g
Casein/meat peptone (50/50)	18.0g
NaCl	5.0g
D-Mannitol	2.5g
Yeast extract	2.0g
Glucose	0.5g
Sodium deoxycholate	0.5g
Brilliant Green	0.01g
Iodine solution	40.0mL
pH 7.5–7.8 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Iodine Solution:**Composition per 40.0mL:**

KI	8.0g
Iodine	5.0g

Preparation of Iodine Solution: Add iodine and KI to distilled/deionized water and bring volume to 40.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 960.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 40.0mL of iodine solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the isolation of *Salmonella* species, except *Salmonella typhi*, and *Arizona* species from food samples.

Tetrathionate Broth, USA (TT Broth, USA)

Na ₂ S ₂ O ₃	30.0g
CaCO ₃	10.0g
Casein peptone	2.5g
Meat peptone	2.5g
Bile salts	1.0g

Source: This medium is available as a premixed powder from Oxoid.

Iodine-Iodide Solution:**Composition per 20.0mL:**

Iodine	6.0g
KI	5.0g

Preparation of Iodine-Iodide Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 20.0mL of iodine solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the selective enrichment of *Salmonella* species from foods.

Tetrathionate Broth with Novobiocin

Composition per liter:

Na ₂ S ₂ O ₃	38.0g
CaCO ₃	25.0g
Casein/meat peptone (50/50)	18.0g
NaCl	5.0g
Yeast extract	2.0g
D-Mannitol	0.5g
Glucose	0.5g
Sodium deoxycholate	0.5g
Brilliant Green	0.01g
Novobiocin	4.0mg
Iodine solution	40.0mL

pH 7.5–7.8 at 25°C

Iodine Solution:

Composition per 40.0mL:

KI	8.0g
Iodine	5.0g

Preparation of Iodine Solution: Add iodine and KI to distilled/deionized water and bring volume to 20.0mL. Mix thoroughly.

Preparation of Medium: Add components, except iodine solution, to distilled/deionized water and bring volume to 960.0mL. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 40°C. Add 40.0mL of iodine solution. Mix thoroughly. Distribute into tubes in 10.0mL volumes. Use medium the same day it is prepared.

Use: For the isolation of *Salmonella* species, except *Salmonella typhi*, and *Arizona* species. Novobiocin suppresses the growth of *Proteus* species.

Tetrathionate Crystal Violet Enhancement Broth

Composition per liter:

Potassium tetrathionate	20.0g
Casein/meat peptone (50/50)	8.6g
NaCl	6.4g
Crystal Violet	0.005g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation of *Salmonella* species, except *Salmonella typhi*, and *Arizona* species from food samples.

Tetrazolium Thallium Glucose Agar

Composition per liter:

Agar	14.0g
Beef extract	10.0g
Peptone	10.0g
Glucose solution	100.0mL
2,3,5-Triphenyltetrazolium·HCl solution	10.0mL
Thallous acetate solution	10.0mL

2,3,5-Triphenyltetrazolium·HCl Solution:

Composition per 10.0mL:

2,3,5-Triphenyltetrazolium·HCl	0.1g

Preparation of 2,3,5-Triphenyltetrazolium·HCl Solution: Add 2,3,5-triphenyltetrazolium·HCl to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Autoclave for 7 min at 15 psi pressure–121°C.

Glucose Solution:

Composition per 100.0mL:

Glucose	10.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Thallous Acetate Solution:

Composition per 10.0mL:

Thallous acetate	1.0g

Preparation of Thallous Acetate Solution:

Add thallous acetate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except glucose solution, 2,3,5-triphenyltetrazolium·HCl solution, and thallous acetate solution—to distilled/deionized water and bring volume to 880.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile glucose solution, 2,3,5-triphenyltetrazolium·HCl solution, and thallous acetate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Streptococcus* species.

Tetrazolium Tolerance Agar (TTC Agar)

Composition per liter:

Pancreatic digest of casein	15.0g

Agar	15.0g
Triphenyltetrazolium chloride	10.0g
Papaic digest of soybean meal	5.0g
NaCl	5.0g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of bacteria based upon the ability to tolerate and grow in the presence of tetrazolium. *Streptococcus faecalis* (enterococci) rapidly reduces tetrazolium.

TGE Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Beef extract	6.0g
Glucose	2.0g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enumeration of bacteria by the membrane filter method.

Thermoacidurans Agar

Composition per liter:

Agar	20.0g
Yeast extract	5.0g
Proteose peptone	5.0g
Glucose	5.0g
K ₂ HPO ₄	4.0g

pH 5.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Bacillus thermoacidurans* from food products.

Thermophilic *Bacillus* Medium

Composition per liter:

Peptone	8.0g
Yeast extract	4.0g
NaCl	3.0g

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a variety of thermophilic *Bacillus* species.

Thiamine Assay Medium

Composition per liter:

Glucose	40.0g
Peptone	22.0g
Sodium acetate	15.0g
Vitamin assay casamino acids	5.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
Adenine sulfate	0.02g
FeSO ₄ ·7H ₂ O	0.02g
Guanine·HCl	0.02g
MnSO ₄ ·5H ₂ O	0.02g
NaCl	0.02g
Uracil	0.02g
L-Cystine	0.2mg
p-Aminobenzoic acid	0.2mg
Calcium pantothenate	0.2mg
Niacin	0.2mg
Pyridoxine·HCl	0.2mg
Riboflavin	0.2mg
Folic acid	0.5μg
Biotin	0.8μg

pH 6.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes or flasks in 5.0mL volumes while swirling the flask to disperse the precipitate. Add standard solutions or test solutions and bring volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the microbiological assaying of thiamine using *Lactobacillus fermentum* as the test organism.

Thiamine Assay Medium LV**Composition per liter:**

Glucose	20.0g
Pancreatic digest of casein	20.0g
K ₂ HPO ₄	10.0g
NaCl	10.0g
Sodium citrate	10.0g
Yeast extract, thiamine-free	10.0g
Sorbitan monooleate complex	2.0g
MgSO ₄ ·7H ₂ O	1.6g
MnCl ₂ ·4H ₂ O	0.28g
FeSO ₄ ·7H ₂ O	0.08g

pH 6.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes or flasks in 5.0mL volumes while swirling the flask to disperse the precipitate. Add standard solutions or test solutions and bring volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the microbiological assaying of thiamine using *Lactobacillus viridescens* as the test organism.

Thiogel® Medium**Composition per liter:**

Gelatin	50.0g
Pancreatic digest of casein	17.0g
Glucose	6.0g
Papaic digest of soybean meal	3.0g
NaCl	2.5g
Sodium thioglycolate	0.5g
Agar	0.7g
Na ₂ SO ₃	0.1g
L-Cystine	0.25g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water preheated to 50°C and bring volume to 1.0L. Mix thoroughly. Let stand for 5 min. Gently heat while stirring and bring to boiling. Distribute into tubes, filling them half full. Autoclave for 15 min at 13 psi pressure–118°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms based on their ability to liquefy gelatin.

Thioglycolate Broth USP, Alternative**Composition per liter:**

Pancreatic digest of casein	15.0g
Glucose	5.5g
Yeast extract	5.0g
NaCl	2.5g
L-Cystine	0.5g
Sodium thioglycolate	0.5g

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Prepare freshly or boil and cool the medium just before use.

Use: For the cultivation of both aerobic and anaerobic organisms in the performance of sterility tests of turbid or viscous specimens.

Thioglycolate Gelatin Medium**Composition per liter:**

Gelatin	50.0g
Pancreatic digest of casein	15.0g
Yeast extract	5.0g
NaCl	2.5g
Glucose	2.0g
Agar	0.75g
L-Cystine	0.25g
Na ₂ SO ₃	0.1g
Thioglycollic acid	0.3mL

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to 50°C. Let stand 5 min. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the determination of gelatin liquefaction by aerobes, microaerophiles, and anaerobes without special incubation.

**Thioglycolate Medium
(DSMZ Medium 530)****Composition per liter:**

Agar	15.0g
Peptone from casein	5.0g
Meat extract	3.0g
Na-thioglycolate solution	100.0mL

pH 5.5 ± 0.2 at 25°C

Thioglycolate Solution :**Composition** per 100mL:

Na-Thioglycolate 0.5g

Preparation of Thioglycolate Solution: Add thioglycolate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except thioglycolate solution to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 100.0mL warm thioglycolate solution. Adjust pH to 5.5. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of various anaerobic bacteria.

Thioglycolate Medium, Brewer**Composition** per liter:

Glucose 5.0g
 Peptone.....5.0g
 NaCl.....5.0g
 Yeast extract.....2.0g
 Sodium thioglycolate 1.1g
 Agar 1.0g
 Beef extract.....1.0g
 Methylene Blue.....2.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For determination of the sterility of solutions containing mercurial preservatives.

**Thioglycolate Medium, Brewer
Modified****Composition** per liter:

Pancreatic digest of casein.....17.5g
 Glucose 10.0g
 NaCl.....5.0g
 Papaic digest of soybean meal.....2.5g
 K₂HPO₄.....2.0g
 Sodium thioglycolate 1.0g
 Agar 0.5g
 Methylene Blue.....0.002g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks, filling them half full. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of obligate anaerobes, microaerophiles, and facultative organisms.

**Thioglycolate Medium, Enriched
(THIO Medium)
(Thioglycolate Medium with
Vitamin K₁ and Hemin)****Composition** per liter:

Thioglycolate medium without indicator..... 1.0L
 Hemin solution..... 0.5mL
 Vitamin K₁ solution 0.1mL

pH 7.0 ± 0.2 at 25°C

Thioglycolate Medium without Indicator:**Composition** per liter:

Pancreatic digest of casein.....17.0g
 Glucose 6.0g
 Papaic digest of soybean meal.....3.0g
 NaCl.....2.5g
 Agar 0.7g
 Sodium thioglycolate 0.5g
 L-Cystine 0.25g
 Na₂SO₃.....0.1g

Source: Thioglycolate medium without indicator is available as a premixed powder from Oxoid and BD Diagnostic Systems.

Preparation of Thioglycolate Medium without Indicator: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Vitamin K₁ Solution:**Composition** per 100.0mL:Vitamin K₁ 1.0g

Preparation of Vitamin K₁ Solution: Add vitamin K₁ to 99.0mL of absolute ethanol. Mix thoroughly.

Hemin Solution:**Composition** per 100.0mL:

Hemin 1.0g
 NaOH (1N solution)..... 20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium: Add 0.5mL of hemin solution and 0.1mL of vitamin K₁ solution to 1.0L of thioglycolate medium without indicator. Mix thoroughly. Distribute into screw-capped tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool tubes or flasks under 85% N₂ + 10% H₂ + 5% CO₂. Tighten caps.

Use: For the isolation, cultivation, and identification of a wide variety of obligate anaerobic bacteria.

**Thioglycolate Medium, Fluid
(Fluid Thioglycolate Medium)
(FTG)
(BAM M146)**

Composition per liter:

Pancreatic digest of casein.....	15.0g
Glucose	5.0g
Yeast extract.....	5.0g
NaCl.....	2.5g
Agar	0.75g
L-cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin solution.....	1.0mL

pH 7.1 ± 0.2 at 25°C

Resazurin Solution:

Composition per 10.0mL:

Na-resazurin.....	10.0mg

Preparation of Resazurin Solution: Add Na-resazurin to 10.0mL of distilled/deionized water. Mix thoroughly. Prepare freshly.

Preparation of Medium: Add components, except sodium thioglycolate and resazurin solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Add 0.5g sodium thioglycolate. Mix thoroughly. Adjust pH to 7.1. Add 1.0mL resazurin solution. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the cultivation of both aerobic and anaerobic organisms in the performance of sterility tests.

Thioglycolate Medium, USP

Composition per liter:

Pancreatic digest of casein.....	15.0g
Glucose	5.5g
Yeast extract.....	5.0g
NaCl.....	2.5g
Agar	0.5g
L-cystine	0.5g
Sodium thioglycolate	0.5g
Resazurin	1.0mg

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of both aerobic and anaerobic organisms in the performance of sterility tests.

**Thioglycolate Medium with 20% Bile
(THIO + Bile Medium)**

Composition per liter:

Oxgall	20.0g
Thioglycolate medium without indicator.....	1.0L
Hemin solution.....	0.5mL
Vitamin K ₁ solution	0.1mL

pH 7.0 ± 0.2 at 25°C

Thioglycolate Medium without Indicator:

Composition per liter:

Pancreatic digest of casein.....	17.0g
Glucose	6.0g
Papaic digest of soybean meal.....	3.0g
NaCl.....	2.5g
Agar	0.7g
Sodium thioglycolate	0.5g
L-Cystine	0.25g
Na ₂ SO ₃	0.1g

Preparation of Thioglycolate Medium without Indicator: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Vitamin K₁ Solution:

Composition per 100.0mL:

Vitamin K ₁	1.0g

Preparation of Vitamin K₁ Solution: Add vitamin K₁ to 99.0mL of absolute ethanol. Mix thoroughly.

Hemin Solution:

Composition per 100.0mL:

Hemin	1.0g
NaOH (1N solution).....	20.0mL

Preparation of Hemin Solution: Add hemin to 20.0mL of 1N NaOH solution. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium: Add 0.5mL of hemin solution, 0.1mL of vitamin K₁ solution, and oxgall to 1.0L of thioglycolate medium without indicator. Mix thoroughly. Distribute into screw-capped tubes or flasks. Autoclave for 15 min at 15 psi pressure–

121°C. Cool tubes or flasks under 85% N₂ + 10% H₂ + 5% CO₂. Tighten caps.

Use: For the isolation, cultivation, and identification of a variety of obligate anaerobic bile-tolerant bacteria.

Thioglycolate Medium without Glucose

Composition per liter:

Pancreatic digest of casein	15.0g
Yeast extract	5.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.25g
Methylene Blue	2.0mg
Thioglycollic acid	0.3mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use (Methylene Blue turns blue), heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of anaerobic, microaerophilic, and aerobic microorganisms. For use in sterility testing of a variety of specimens.

Thioglycolate Medium without Glucose

Composition per liter:

Pancreatic digest of casein	20.0g
NaCl	2.5g
K ₂ HPO ₄	1.5g
Sodium thioglycolate	0.6g
Agar	0.5g
L-Cystine	0.4g
Na ₂ SO ₃	0.2g
Methylene Blue	2.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks, filling them half full. Autoclave for 15 min at 15 psi pressure–121°C.

Use: Use as a base for fermentation studies of anaerobic bacteria and for the promotion of endospore formation.

Thioglycolate Medium without Glucose and Indicator

Composition per liter:

Pancreatic digest of casein	15.0g
Yeast extract	5.0g
NaCl	2.5g
Agar	0.75g
L-Cystine	0.25g
Thioglycollic acid	0.3mL

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use, heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of anaerobic, microaerophilic, and aerobic microorganisms. For use in sterility testing of a variety of specimens.

Thioglycolate Medium without Indicator

Composition per liter:

Pancreatic digest of casein	15.0g
Yeast extract	5.0g
Glucose	5.0g
NaCl	2.5g
Agar	0.75g
Sodium thioglycolate	0.5g
L-Cystine	0.25g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. If medium becomes oxidized before use, heat in a boiling water bath to expel absorbed O₂. Cool to 25°C.

Use: For the cultivation of anaerobic, microaerophilic, and aerobic microorganisms. For use in sterility testing of a variety of specimens.

Thioglycolate Medium without Indicator

Composition per liter:

Pancreatic digest of casein	17.0g
Glucose	6.0g

Papaic digest of soybean meal	3.0g
NaCl	2.5g
Agar	0.7g
Sodium thioglycolate	0.5g
L-Cystine	0.25g
Na ₂ SO ₃	0.1g

pH 7.0 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Prepare freshly or boil and cool the medium just before use.

Use: For the growth of aerobic and anaerobic microorganisms.

TNT Medium

(Tryptone NaCl Thiamine Medium)

Composition per liter:

Pancreatic digest of casein	10.0g
NaCl	5.0g
Thiamine-HCl	1.0mg

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli*.

Todd-Hewitt Broth

Composition per liter:

Pancreatic digest of casein	20.0g
Infusion from 450.0g fat-free minced meat	10.0g
Glucose	2.0g
NaHCO ₃	2.0g
NaCl	2.0g
Na ₂ HPO ₄	0.4g

pH 7.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–115°C.

Use: For the cultivation of group A streptococci used in serological typing, and for the cultivation of a variety of pathogenic microorganisms.

Todd-Hewitt Broth (ATCC Medium 235)

Composition per liter:

Peptone	20.0g
Beef heart, solids from infusion	3.1g
Na ₂ CO ₃	2.5g
Glucose	2.0g
NaCl	2.0g
Na ₂ HPO ₄	0.4g

pH 7.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of Group A streptococci used in serological typing, and for the cultivation of a variety of pathogenic microorganisms.

Todd-Hewitt Broth, Modified

Composition per liter:

Neopeptone	20.0g
Glucose	2.0g
NaHCO ₃	2.0g
NaCl	2.0g
Na ₂ HPO ₄	0.4g
Beef heart infusion	1.0L

pH 7.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 10 psi pressure–115°C.

Use: For the cultivation of streptococci for serological identification.

Toluidine Blue DNA Agar (BAM M148)

Composition per liter:

Agar	10.0g
NaCl	10.0g
Tris(hydroxymethyl)aminomethane buffer	6.1g
Deoxyribonucleic acid	0.3g
Toluidine Blue O	0.083g
CaCl ₂ , anhydrous	1.1mg

pH 9.0 ± 0.2 at 25°C

Preparation of Medium: Add tris(hydroxymethyl)aminomethane buffer to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 9.0. Add the remaining components, except Toluidine Blue O. Mix thoroughly. Gently heat and bring to boiling. Add Toluidine Blue O. Mix thor-

oughly. If used the same day, sterilization is not necessary. Cool to 50°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and differentiation of *Staphylococcus aureus* from foods.

Toluidine Blue DNA Agar

Composition per liter:

Agar	10.0g
NaCl	10.0g
Tris(hydroxymethyl)aminomethane buffer	6.1g
Deoxyribonucleic acid (DNA)	0.3g
Toluidine Blue O	0.083g
CaCl ₂ , anhydrous	1.1mg
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except Toluidine Blue O, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Add Toluidine Blue O. Mix thoroughly. Medium does not have to be sterilized if used immediately. Pour into sterile Petri dishes or distribute into sterile tubes. Allow tubes to cool in a slanted position.

Use: For the cultivation and differentiation of bacteria based on their production of deoxyribonuclease (DNase). Bacteria that produce DNase turn the medium pink.

Tomato Juice Agar

Composition per liter:

Agar	12.0g
Pancreatic digest of casein	10.0g
Peptonized milk	10.0g
Tomato juice	400.0mL
pH 6.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of lactobacilli, especially *Lactobacillus acidophilus*.

Tomato Juice Agar (ATCC Medium 33)

Composition per liter:

Agar	11.0g
Pancreatic digest of casein	10.0g

Yeast extract	10.0g
Tomato juice, filtered	200.0mL
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a variety of bacteria including *Lactobacillus*, *Leuconostoc*, *Pediococcus*, and *Propionibacterium* species.

Tomato Juice Agar Special

Composition per liter:

Agar	20.0g
Pancreatic digest of casein	10.0g
Peptonized milk	10.0g
Tomato juice	400.0mL
pH 5.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Avoid overheating—it results in a soft agar. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of lactobacilli.

Tomato Juice Broth

Composition per liter:

Tomato juice, dessicated	20.0g
Glucose	10.0g
Yeast extract	10.0g
K ₂ HPO ₄	0.5g
KH ₂ PO ₄	0.5g
MgSO ₄ ·7H ₂ O	0.2g
FeSO ₄ ·7H ₂ O	0.01g
MnSO ₄ ·7H ₂ O	0.01g
NaCl	0.01g
pH 6.7 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of yeast and other aciduric microorganisms.

Tomato Juice Broth (ATCC Medium 433)

Composition per liter:

Pancreatic digest of casein	10.0g
Yeast extract	10.0g
Tomato juice, filtered	200.0mL
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a variety of fastidious bacteria that require complex growth factors, including *Lactobacillus*, *Aerococcus*, *Bifidobacterium*, and *Pediococcus* species.

Tomato Juice Medium

Composition per liter:

Agar	15.0g
Glucose	10.0g
Polypeptone	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	0.5g
CaCl ₂ ·2H ₂ O	0.125g
KCl	0.125g
MgSO ₄ ·7H ₂ O	0.125g
NaCl	0.125g
Bromocresol Green	0.03g
MnSO ₄ ·4H ₂ O	3.0mg
Tomato juice, canned	150.0mL
Cycloheximide solution	10.0mL
pH 5.0 ± 0.2 at 25°C	

Cycloheximide Solution:

Composition per 10.0mL:

Cycloheximide	0.1g

Preparation of Cycloheximide Solution: Add cycloheximide to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloheximide solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile cycloheximide solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Lactobacilli* from wine.

Tomato Juice Medium

Composition per liter:

Tryptic digest of casein	10.0g
Yeast extract	10.0g
Tomato juice, filtered, pH 7.0	200.0mL
Tween 80	1.0mL
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Lactobacillus collinoides*.

Tomato Juice Milk Agar (DSMZ Medium 353)

Composition per liter:

Skim Milk	100.0g
Yeast extract	5.0g
Tomato juice	100.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Filter canned tomatoes through paper to produce tomato juice. Leave over night at 10°C. Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Enterococcus faecalis*=*Streptococcus faecalis*.

Tomato Juice Yeast Extract Medium

Composition per liter:

Skim milk	100.0g
Yeast extract	5.0g
Tomato juice, filtered	100.0mL

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of yeasts and fungi.

Tomato Juice Yeast Extract Milk Medium

Composition per liter:

Skim milk	100.0g
Yeast extract	5.0g
Tomato juice, filtered	100.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Filter canned tomato juice through paper. Let stand overnight at 10°C. Add

remaining components and bring to 1.0L with distilled/deionized water. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a variety of fastidious bacteria that require complex growth factors, including *Lactobacillus*, *Streptococcus*, and *Enterococcus* species.

TPEY Agar

(Tellurite Polymyxin Egg Yolk Agar)

Composition per liter:

NaCl.....	20.0g
Agar	15.5g
Pancreatic digest of casein.....	10.0g
Yeast extract.....	5.0g
D-Mannitol	5.0g
LiCl	2.0g
Egg yolk emulsion (30% solution)	100.0mL
Chapman tellurite solution.....	10.0mL
Polymyxin B solution	0.4mL
pH 7.1 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Egg Yolk Emulsion (30% Solution):

Composition per 100.0mL:

NaCl.....	0.6g
Egg yolk.....	30.0mL

Preparation of Egg Yolk Emulsion (30% Solution): Add NaCl and egg yolk to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Chapman Tellurite Solution:

Composition per 100.0mL:

K ₂ TeO ₃	1.0g

Preparation of Chapman Tellurite Solution: Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Polymyxin B Solution:

Composition per 100.0mL:

Polymyxin B	1.0g

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components—except 30% egg yolk emulsion, Chapman tellurite solution, and polymyxin B solution—to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to

45°–50°C. Aseptically add 100.0mL of sterile 30% egg yolk emulsion, 10.0mL of sterile Chapman tellurite solution, and 0.4mL of sterile polymyxin B solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the recovery of staphylococci from foods and other materials.

TPGY Medium

(Thioglycolate Peptone

Glucose Yeast Extract Medium)

Composition per liter:

Pancreatic digest of casein.....	50.0g
Peptone	5.0g
Yeast extract.....	5.0g
Glucose	1.0g
Sodium thioglycolate	1.0g
pH 7.1 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of anaerobic bacteria.

Tributyryn Agar

Composition per liter:

Agar	15.0g
Tributyryn (glyceryl tributyrate).....	10.0g
Peptone	5.0g
Yeast extract.....	3.0g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a prepared medium from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and enumeration of lipolytic fungi and bacteria, especially *Staphylococcus* species, *Flavobacterium* species, *Clostridium* species, and *Pseudomonas* species from butter. Lipolytic bacteria appear as colonies surrounded by a clear zone.

Trimethylamine N-Oxide Medium (TMAO Medium)

Composition per liter:

Beef extract.....	10.0g
Peptone	10.0g

NaCl.....	5.0g
Agar	2.0g
Trimethylamine N-oxide.....	1.0g
Yeast extract.....	1.0g
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped tubes in 4.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in an upright position.

Use: For the cultivation and differentiation of *Campylobacter* species from foods. *Campylobacter jejuni* and *Campylobacter coli* will not grow.

Triple Sugar Iron Agar (TSI Agar)

Composition per liter:

Peptone.....	20.0g
Agar	12.0g
Lactose.....	10.0g
Sucrose.....	10.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Yeast extract.....	3.0g
Glucose	1.0g
Ferric citrate.....	0.3g
Na ₂ S ₂ O ₃	0.3g
Phenol Red.....	0.025g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position to form a 1.0-inch butt.

Use: For the differentiation of members of the *Enterobacteriaceae* based on their fermentation of lactose, sucrose, and glucose and the production of H₂S.

Triple Sugar Iron Agar (TSI Agar) (BAM M149 Medium 1)

Composition per liter:

Polypeptone	20.0g
Agar	13.0g
Lactose.....	10.0g
Sucrose.....	10.0g

NaCl.....	5.0g
Glucose	1.0g
Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O	0.2g
Na ₂ S ₂ O ₃	0.2g
Phenol Red.....	0.025g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for min at 13 psi pressure–118°C. Allow tubes to cool in a slanted position to form a 1.0-inch butt.

Use: For the differentiation of members of the *Enterobacteriaceae* based on their fermentation of lactose, sucrose, and glucose and the production of H₂S.

Triple Sugar Iron Agar (TSI Agar) (BAM M149 Medium 2)

Composition per liter:

Peptone	15.0g
Agar	12.0g
Lactose.....	10.0g
Sucrose.....	10.0g
Proteose peptone.....	5.0g
NaCl.....	5.0g
Beef extract.....	3.0g
Yeast extract.....	3.0g
Glucose	1.0g
Na ₂ S ₂ O ₃	0.3g
FeSO ₄	0.2g
Phenol Red.....	0.024g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position to form a 1.0-inch butt.

Use: For the differentiation of members of the *Enterobacteriaceae* based on their fermentation of lactose, sucrose, and glucose and the production of H₂S.

Triple Sugar Iron Agar (TSI Agar)

Composition per liter:

Agar	13.0g
Pancreatic digest of casein.....	10.0g
Peptic digest of animal tissue	10.0g
Lactose.....	10.0g
Sucrose.....	10.0g
NaCl.....	5.0g

Glucose	1.0g
Fe(NH ₄) ₂ (SO ₄) ₂ ·6H ₂ O	0.2g
Na ₂ S ₂ O ₃	0.2g
Phenol Red	0.025g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position to form a 1.0 inch butt.

Use: For the differentiation of members of the Enterobacteriaceae based on their fermentation of lactose, sucrose, and glucose and the production of H₂S.

Trypaflavin Nalidixic Acid Serum Agar (TNSA Agar)

Composition per liter:

Ionagar No. 2	12.0g
Peptone.....	10.0g
Beef extract.....	3.0g
H ₂ O	926.5mL
Bovine serum, heat inactivated.....	50.0mL
Nalidixic acid solution.....	20.0mL
Trypaflavin solution.....	3.5mL
pH 7.2–7.4 at 25°C	

Nalidixic Acid Solution:

Composition per 10.0mL:

Nalidixic acid.....	0.02g

Preparation of Nalidixic Acid Solution: Add nalidixic acid to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Trypaflavin Solution:

Composition per 10.0mL:

Trypaflavin.....	0.1g

Preparation of Trypaflavin Solution: Add trypaflavin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components—except bovine serum, nalidixic acid solution, and trypaflavin solution—to distilled/deionized water and bring volume to 926.5mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile bovine serum, 20.0mL of sterile nalidixic acid solution, and 3.5mL of trypaflavin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Listeria* species from preenriched specimens.

Tryptic Nitrate Medium

Composition per liter:

Tryptose	20.0g
Na ₂ HPO ₄	2.0g
Agar	1.0g
Glucose	1.0g
KNO ₃	1.0g
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of *Pseudomonas* and related genera. For the differentiation of bacteria based on their reduction of nitrate to nitrite. After incubation of the bacterium in tryptic nitrate medium for 18–24 hr, sulfanilic acid and α -naphthol reagents are added. Nitrate reduction is indicated by the development of a red to violet color.

Tryptic Soy Agar with Magnesium Sulfate

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	15.0g
NaCl.....	5.0g
Pancreatic digest of soybean meal.....	5.0g
MgSO ₄ ·7H ₂ O	1.5g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Escherichia coli* from foods.

Tryptic Soy Agar with Magnesium Sulfate and Sodium Chloride

Composition per liter:

Pancreatic digest of casein.....	50.0g
NaCl.....	30.0g
Agar	15.0g
Pancreatic digest of soybean meal.....	5.0g
MgSO ₄ ·7H ₂ O	1.5g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Vibrio* species from foods.

Tryptic Soy Agar with Sodium Chloride (ATCC Medium 2276)

Composition per liter:

Pancreatic digest of casein.....	50.0g
NaCl.....	20.0g
Agar	15.0g
Pancreatic digest of soybean meal.....	5.0g
MgSO ₄ ·7H ₂ O	1.5g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Vibrio* species.

Tryptic Soy Agar with 0.6% Yeast Extract

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	15.0g
Yeast extract.....	6.0g
Pancreatic digest of soybean meal.....	5.0g
NaCl.....	5.0g
pH 7.0–7.5 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Listeria monocytogenes* from foods.

Tryptic Soy Fast Green Agar (TSFA)

Composition per liter:

Pancreatic digest of casein.....	17.0g
Agar	15.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g

K ₂ HPO ₄	2.5g
Glucose	2.5g
Fast Green FCF.....	0.25g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically adjust pH to 7.3. Pour into sterile Petri dishes.

Use: For the isolation and cultivation of *Salmonella* species from foods.

Trypticase Agar Base

Composition per liter:

Pancreatic digest of casein.....	20.0g
Agar	3.5g
Phenol Red.....	0.02g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differentiation of microorganisms based on their motility.

Trypticase Agar Base with Carbohydrate

Composition per liter:

Pancreatic digest of casein.....	20.0g
Carbohydrate.....	5.0g
Agar	3.5g
Phenol Red.....	0.02g
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 13 psi pressure–118°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For differentiation of microorganisms based on their motility and fermentation reactions. Fermentation of carbohydrate changes the medium yellow.

Trypticase Glucose Extract Agar

Composition per liter:

Agar	15.0g

Pancreatic digest of casein	5.0g
Beef extract	3.0g
Glucose	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of bacteria in water, milk, and other specimens.

Trypticase Novobiocin Broth (TN Broth) (BAM M150)

Composition per liter:

Pancreatic digest of casein	17.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g
Bile salts No. 3	1.5g
K ₂ HPO ₄	1.5g
Novobiocin solution	10.0mL
pH 7.3 ± 0.2 at 25°C	

Novobiocin Solution:

Composition per 10.0mL:

Novobiocin	0.02g

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile novobiocin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of verotoxin-producing *Escherichia coli*.

Trypticase Peptone Glucose Yeast Extract Broth (TPGY Broth) (BAM M151)

Composition per liter:

Pancreatic digest of casein	50.0g
Yeast extract	20.0g

Peptone	5.0g
Glucose	4.0g
Sodium thioglycolate	1.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 15.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the cultivation of *Clostridium botulinum*.

Trypticase Peptone Glucose Yeast Extract Broth, Buffered

Composition per liter:

Pancreatic digest of casein	50.0g
Yeast extract	20.0g
Na ₂ HPO ₄	5.0g
Peptone	5.0g
Glucose	4.0g
Sodium thioglycolate	1.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 7.3. Distribute into tubes in 15.0mL volumes. Autoclave for 8 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Clostridium perfringens* from foods.

Trypticase Peptone Glucose Yeast Extract Broth with Trypsin (TPGYT Broth) (BAM M151a)

Composition per 1067.0mL:

Pancreatic digest of casein	50.0g
Yeast extract	20.0g
Peptone	5.0g
Glucose	4.0g
Sodium thioglycolate	1.0g
Trypsin solution	67.0mL
pH 7.0 ± 0.2 at 25°C	

Trypsin Solution:

Composition per 100.0mL:

Trypsin	1.5g

Preparation of Trypsin Solution: Add trypsin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except trypsin solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 15.0mL

volumes. Autoclave for 10 min at 15 psi pressure–121°C. Immediately prior to use, aseptically add 1.0mL of sterile trypsin solution to each tube. Mix thoroughly.

Use: For the cultivation of *Clostridium botulinum*.

Trypticase Soy Agar

Composition per liter:

Pancreatic digest of casein.....	17.0g
Agar	15.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of heterotrophic microorganisms.

Trypticase Soy Agar (ATCC Medium 18)

Composition per liter:

Pancreatic digest of casein.....	17.0g
Agar	15.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of fastidious and nonfastidious microorganisms from clinical and nonclinical specimens. Also used for the rapid estimation of the bacteriological quality of water.

Trypticase Soy Agar (Tryptic Soy Agar) (Soybean Casein Digest Agar) (ATCC Medium 77) (BAM M152)

Composition per liter:

Pancreatic digest of casein.....	15.0g
Agar	15.0g

Papaic digest of soybean meal.....	5.0g
NaCl.....	5.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of a wide variety of fastidious as well as nonfastidious microorganisms.

Trypticase Soy Agar with Lecithin and Polysorbate 80 (Microbial Content Test Agar)

Composition per liter:

Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g
NaCl.....	5.0g
Polysorbate 80 (Tween 80).....	5.0g
Lecithin.....	0.7g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 13 psi pressure–118°C. Cool to 45°–50°C. Pour into sterile Petri dishes in 17.0mL volumes or leave in tubes.

Use: For the detection and enumeration of microorganisms in replicate plating techniques. Also used for the detection and enumeration of microorganisms present on surfaces of sanitary importance.

Trypticase Soy Agar-Magnesium Sulfate-NaCl Agar (TSAMS) (BAM M152a)

Composition per liter:

NaCl.....	20.0g
Trypticase peptone.....	15.0g
Agar	15.0g
Phytone peptone.....	5.0g
MgSO ₄ ·7H ₂ O	1.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes in 20.0mL volumes.

Use: For the cultivation of *Vibrio* species from foods

Trypticase Soy Agar with 3% NaCl (TSA NaCl)

Composition per liter:

NaCl.....	30.0g
Agar	15.0g
Pancreatic digest of casein.....	15.0g
Papaic digest of soybean meal.....	5.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of halophilic microorganisms isolated from foods.

Trypticase Soy Agar with NaCl (ATCC Medium 176)

Composition per liter:

NaCl.....	30.0g
Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g
Bile salts No. 3.....	1.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Vibrio alginolyticus*.

Trypticase Soy Agar with NaCl, Horse Serum, and Penicillin

Composition per liter:

NaCl.....	35.0g
Pancreatic digest of casein.....	15.0g
Agar	15.0g
Papaic digest of soybean meal.....	5.0g
Horse serum, inactivated.....	100.0mL
Penicillin solution	10.0mL
pH 7.3 ± 0.2 at 25°C	

Penicillin Solution:

Composition per 10.0mL:

Penicillin.....	1,000,000U

Preparation of Penicillin Solution: Add penicillin to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except horse serum and penicillin solution, to distilled/deionized water and bring volume to 890.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Cool to 50°C. Aseptically add 100.0mL of sterile horse serum and 10.0mL of sterile penicillin solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of fungi.

Trypticase Soy Agar Yeast Extract (TSAYE) (BAM M153)

Composition per liter:

Pancreatic digest of casein.....	17.0g
Agar	15.0g
Yeast extract.....	6.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of heterotrophic microorganisms. For the isolation and cultivation of *Listeria monocytogenes* from foods.

Trypticase Soy Broth (Soybean Casein Digest Broth, USP) (Tryptic Soy Broth) (LMG Medium 185) (BAM M154)

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g

K ₂ HPO ₄	2.5g
Glucose	2.5g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of microorganisms.

Trypticase Soy Broth, Modified (mTSB) (BAM M156)

Composition per 1000.2mL:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
K ₂ HPO ₄	4.0g
Papaic digest of soybean meal.....	3.0g
Glucose	2.5g
Bile salts No. 3.....	1.5g
Novobiocin solution.....	0.2mL
pH 7.3 ± 0.2 at 25°C	

Novobiocin Solution:

Composition per liter:

Novobiocin.....	0.05g

Preparation of Novobiocin Solution: Add novobiocin to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except novobiocin solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile novobiocin solution. Mix thoroughly. Aseptically distribute into sterile tubes.

Use: For the isolation and cultivation of *Shigella* species from food.

Trypticase Soy Broth with 10 mM Glucose (ATCC Medium 1189)

Composition per liter:

Pancreatic digest of casein.....	18.0g
Papaic digest of soybean meal.....	6.0g
NaCl.....	6.0g
Glucose	1.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium without glucose is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Staphylococcus aureus* subsp. *Aureus*

Trypticase Soy Broth with Ferrous Sulfate

(Tryptic Soy Broth with Ferrous Sulfate) (BAM M186)

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g
FeSO ₄	35.0mg
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 7.3. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the detection of *Salmonella* spp. from foods.

Trypticase Soy Broth with Glycerol (Tryptic Soy Broth with Glycerol) (TSB) (BAM M155)

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glycerol	240.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 7.3. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a wide variety of microorganisms from foods. Used to grow bacteria for cryopreservation.

Trypticase Soy Broth with Sodium Chloride and Sodium Pyruvate (BAM M154a)

Composition per liter:

NaCl.....	100.0g
Pancreatic digest of casein.....	17.0g
Sodium pyruvate.....	10.0g
Papaic digest of soybean meal.....	3.0g
Glucose.....	2.5g
K ₂ HPO ₄	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 7.3. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Staphylococcus aureus* from foods.

Trypticase Soy Broth with Yeast Extract (BAM M157)

Composition per liter:

Pancreatic digest of casein.....	17.0g
Yeast extract.....	6.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose.....	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of a wide variety of heterotrophic microorganisms.

Trypticase Soy Broth without Glucose

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of microorganisms when the presence of carbohydrate is undesirable.

Trypticase Soy Broth Yeast Extract (TSBYE)

Composition per liter:

Pancreatic digest of casein.....	17.0g
Yeast extract.....	6.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose.....	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Listeria monocytogenes* from foods.

Trypticase Soy Polymyxin Broth (BAM M158)

Composition per 1006.67mL:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Papaic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose.....	2.5g
Polymyxin B solution.....	6.67mL

pH 7.3 ± 0.2 at 25°C

Polymyxin B Solution:

Composition per 10.0mL:

Polymyxin B.....	0.015g

Preparation of Polymyxin B Solution: Add polymyxin B to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except polymyxin B solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes in 15.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 0.1mL of sterile polymyxin B solution to each tube. Mix thoroughly.

Use: For the isolation and cultivation of *Bacillus cereus* from foods.

Trypticase Soy-Sheep Blood Agar
(Tryptic Soy Blood Agar)
(TSA Blood Agar)
(BAM M159)

Composition per 1050mL:

Pancreatic digest of casein	15.0g
Agar	15.0g
Papaic digest of soybean meal	5.0g
NaCl	5.0g
Sheep blood, defibrinated	50.0mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL sterile sheep blood. Mix thoroughly. Pour into sterile Petri dishes in 20.0mL volumes or distribute into sterile tubes.

Use: For the cultivation of a wide variety of fastidious microorganisms. For the observation of hemolytic reactions of a variety of bacteria. May be used to perform the CAMP test for the presumptive identification of group B streptococci (*Streptococcus agalactiae*).

Trypticase Soy Tryptose Broth
(BAM M160)

Composition per liter:

Pancreatic digest of casein	13.5g
Peptic digest of animal tissue	5.0g
NaCl	5.0g
Yeast extract	3.0g
Glucose	1.75g
Papaic digest of soybean meal	1.5g
K ₂ HPO ₄	1.25g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enrichment of *Salmonella* species from foods.

Trypticase Soy Yeast Extract Agar

Composition per liter:

Agar	28.0g
NaCl	10.0g
Pancreatic digest of casein	2.5g
Yeast extract	2.5g

Base medium	100.0mL
Phosphate buffer	100.0mL
pH 7.2 ± 0.2 at 25°C	

Base Medium:**Composition per liter:**

MgCl ₂ ·6H ₂ O	2.0g
Titriplex I	1.0g
CaSO ₄ ·2H ₂ O	0.4g
NaOH-pellets	0.2g
Fe-citrate solution	5.0mL
Trace elements solution	5.0mL

Fe-Citrate Solution:**Composition per 100.0mL:**

Fe-citrate	0.25g

Preparation of Fe-Citrate Solution: Add Fe-citrate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Trace Elements Solution:**Composition per 5.0mL:**

CuCl ₂ ·2H ₂ O	50.0mg
Titriplex I	12.8mg
FeCl ₂ ·4H ₂ O	1.0mg
MnCl ₂ ·4H ₂ O	0.5mg
CoCl ₂ ·6H ₂ O	0.3mg
ZnCl ₂	0.2mg
Na ₂ MoO ₄ ·2H ₂ O	50.0ng
NiCl ₂ ·6H ₂ O	50.0ng
H ₃ BO ₃	20.0ng

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 5.0mL. Mix thoroughly.

Preparation of Base Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Phosphate Buffer:**Composition per liter:**

Na ₂ HPO ₄ ·12H ₂ O	43.0g
KH ₂ PO ₄	5.44g

Preparation of Phosphate Buffer: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C.

Preparation of Medium: Add components, except phosphate buffer, and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile phosphate buffer. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Lactococcus lactis* and *Lactococcus plantarum*.

Trypticase Yeast Extract Glucose Medium

Composition per liter:

Pancreatic digest of casein	10.0g
Glucose	10.0g
KH ₂ PO ₄	6.8g
Yeast extract	5.0g
NaHCO ₃	1.0g
Tween 80	0.5g
Sodium formaldehyde sulfoxalate	0.5g
CaCl ₂	0.02g
MgSO ₄	0.02g
NaCl	0.02g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Propionibacterium* species.

Tryptone Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	8.0g
NaCl	8.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms such as *Escherichia coli* and *Pseudomonas* species.

Tryptone Bile Agar

Composition per liter:

Pancreatic digest of casein	20.0g
Agar	15.0g
Bile salts No. 3	1.5g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the isolation and enumeration of *Escherichia coli* from foods.

Tryptone Bile Glucuronide Agar, Harlequin (Harlequin TBGA)

Composition per liter:

Tryptone	20.0g
Agar	15.0g
Bile salts No. 3	1.5g
X-glucuronide	0.075g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available from lab m.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Allow to soak for 10 min. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the simple enumeration of *E. coli* without the need for membranes, or pre-incubation. The medium has been modified by the addition of a chromogenic substrate to detect the β-glucuronidase, which is highly specific for *E. coli*. The advantage of the chromogenic substrate is that it requires no UV lamp to visualize the reaction, and it is concentrated within the colony, facilitating easier enumeration in the presence of other organisms, or when large numbers are present on the plate.

Tryptone Bile X-glucuronide Agar, Chromocult (Chromocult TBX) (Chromocult Tryptone Bile X-glucuronide Agar)

Composition per liter:

Peptone	20.0g
Agar	15.0g
Bile salts No. 3	1.5g
X-β-D-glucuronide	0.075g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the differentiation of *E. coli* from other coliforms. The presence of the enzyme β-D-glucuronidase differentiates most *E. coli* spp. from other coliforms. *E. coli* absorbs the chromogenic substrate 5-bromo-4-chloro-3-indolyl-β-D-glucuronide (X-β-D-glucuronide). The enzyme β-glucuronidase splits the bond between the chromophore 5-bromo-4-chloro-3-indolyl- and the β-D-glucuronide. *E. coli* colonies are

colored blue-green. Growth of accompanying Gram-positive flora is largely inhibited by the use of bile salts. The prepared medium is clear and yellowish.

Tryptone Broth

Composition per liter:

Pancreatic digest of casein.....	10.0g
Glucose	5.0g
K ₂ HPO ₄	1.25g
Yeast extract.....	1.0g
Bromcresol Purple solution	2.0mL

Bromcresol Purple Solution:

Composition per 100.0mL:

Bromcresol Purple	2.0g
Ethanol	10.0mL

Preparation of Bromcresol Purple Solution:

Add Bromcresol Purple to 10.0mL of ethanol. Mix thoroughly. Bring volume to 100.0mL with distilled/deionized water.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped tubes in 10.0mL volumes. Autoclave for 20 min at 15 psi pressure–121°C.

Use: For the cultivation of *Salmonella* species from foods.

Tryptone Broth (ATCC Medium 274) (BAM M164)

Composition per liter:

Pancreatic digest of casein.....	10.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms such as *Escherichia coli* and *Pseudomonas* species.

Tryptone Broth with CaCl₂

Composition per liter:

Pancreatic digest of casein.....	10.0g
CaCl ₂	5.5g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms such as *Escherichia coli*.

Tryptone Broth, 1% (ATCC Medium 274)

Composition per liter:

Pancreatic digest of casein.....	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli* and *Pseudomonas* species. For the identification and confirmation of *Vibrio cholerae* in foods.

Tryptone Glucose Beef Extract Agar (Tryptone Glucose Extract Agar)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	5.0g
Beef extract.....	3.0g
Glucose	1.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. If the dilution of the specimen is greater than 1:10, add 10.0mL of sterile 10% skim milk solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of bacteria by the standard plate count procedure. For the cultivation and enumeration of bacteria from milk and dairy products. For the detection of thermophilic organisms.

Tryptone Glucose Beef Extract Agar with Sucrose

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	5.0g
Beef extract.....	3.0g
Glucose	1.0g
Sucrose.....	5.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharococcus thermophilus*.

Tryptone Phosphate Brain Heart Infusion Yeast Extract Agar (TPBY)

Composition per liter:

Pancreatic digest of casein	20.0g
Agar	15.0g
NaCl	5.14g
K ₂ HPO ₄	2.0g
KH ₂ PO ₄	2.0g
Yeast extract	1.0g
Oxgall	0.5g
Pancreatic digest of gelatin	0.4g
Brain heart, solids from infusion	0.16g
Peptic digest of animal tissue	0.16g
Glucose	0.08g
Na ₂ HPO ₄	0.06g
Tween 80	1.5mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of coliform bacteria, such as *Escherichia coli*, from foods.

Tryptone Phosphate Broth (BAM M162)

Composition per liter:

Pancreatic digest of casein	20.0g
NaCl	5.0g
K ₂ HPO ₄	2.0g
KH ₂ PO ₄	2.0g
Tween 80	15.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of enteropathogenic *Escherichia coli*.

Tryptone Phosphate Broth

Composition per liter:

Pancreatic digest of casein	20.0g
NaCl	5.0g
K ₂ HPO ₄	2.0g
KH ₂ PO ₄	2.0g
Tween 80	1.5mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of coliform bacteria, such as *Escherichia coli*, from foods.

Tryptone Soya Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	15.0g
NaCl	5.0g
Pancreatic digest of soybean meal	5.0g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of microorganisms.

Tryptone Soya Agar pH 5.5 (LMG Medium 241)

Composition per liter:

Tryptone	15.0g
Agar	15.0g
Soya peptone	5.0g
NaCl	5.0g

pH 5.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of heterotrophic bacteria.

Tryptone Soya Agar pH 6.5 (LMG Medium 242)

Composition per liter:

Tryptone.....	15.0g
Agar.....	15.0g
Soya peptone.....	5.0g
NaCl.....	5.0g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of heterotrophic bacteria.

Tryptone Soya Agar with 4.5% NaCl

Composition per liter:

NaCl.....	45.0g
Pancreatic digest of casein.....	17.0g
Agar.....	12.0g
Papaic digest of soybean meal.....	3.0g
Glucose.....	2.5g
K ₂ HPO ₄	2.5g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Brevibacterium linens*.

Tryptone Soya Agar with Sheep Blood (LMG Medium 175)

Composition per liter:

Agar.....	15.0g
Pancreatic digest of casein.....	15.0g
NaCl.....	5.0g
Pancreatic digest of soybean meal.....	5.0g
Sheep blood, defibrinated.....	50.0mL

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Heat with frequent agitation and boil for 1 min to completely dissolve. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 50.0mL of sterile, defibrinated sheep blood. Mix thoroughly and pour into sterile Petri dishes.

Use: For cultivation and maintenance of *Bacillus* spp.

Tryptone Soya Broth

Composition per liter:

Pancreatic digest of casein.....	17.0g
NaCl.....	5.0g
Pancreatic digest of soybean meal.....	3.0g
K ₂ HPO ₄	2.5g
Glucose.....	2.5g

pH 7.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of microorganisms.

Tryptone Water Broth (Tryptone Broth)

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	5.0g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Dissolve 15.0g in 1.0L of distilled water and distribute into final containers. Sterilize by autoclaving at 121°C for 15 min.

Use: For the cultivation of production of indole by microorganisms.

Tryptone with NaCl Broth

Composition per liter:

Pancreatic digest of casein.....	8.0g
NaCl.....	0.5g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms such as *Escherichia coli* and *Pseudomonas* species.

Tryptone Yeast Extract Agar (BAM M165)

Composition per liter:

Pancreatic digest of casein.....	10.0g
Agar.....	2.0g

Yeast extract.....	1.0g
Bromcresol Purple	0.04g
Carbohydrate solution.....	100.0mL
pH 7.0 ± 0.2 at 25°C	

Carbohydrate Solution:**Composition** per 100.0mL:

Carbohydrate.....	10.0g

Preparation of Carbohydrate Solution: Add carbohydrate (glucose or mannitol for *Staphylococcus aureus*) to distilled/deionized water and bring volume to 100.0mL. Glucose or mannitol may be used. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Adjust pH to 7.0. Gently heat and bring to boiling. Distribute into tubes in 13.5mL volumes. Autoclave for 20 min at 10 psi pressure–115°C. Cool to 45°–50°C. Aseptically add 1.5mL of carbohydrate solution to each tube. Mix thoroughly. Solidify agar quickly by placing tubes in ice water.

Use: For the cultivation and differentiation of *Staphylococcus aureus* based on glucose and mannitol fermentation. Bacteria that ferment the added carbohydrate turn the medium yellow.

Tryptone Yeast Extract Agar 1**Composition** per liter:

Agar	20.0g
Pancreatic digest of casein.....	10.0g
Yeast extract.....	5.0g
K ₂ HPO ₄	4.4g
Glucose	2.0g
NaCl.....	2.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of heterotrophic bacteria.

Tryptophan 1% Solution
(Trypticase 1% Solution)
(Tryptone 1% Solution)

Composition per liter:

Pancreatic digest of casein.....	10.0g
pH 7.0 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of bacteria, especially members of the *Enterobacteriaceae*, based on their production of indole.

Tryptophan Assay Medium**Composition** per liter:

Glucose	40.0g
Sodium acetate.....	20.0g
Casamino acids	12.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
L-Cystine	0.2g
Adenine sulfate	0.02g
FeSO ₄ ·7H ₂ O.....	0.02g
Guanine·HCl.....	0.02g
MnSO ₄ ·7H ₂ O	0.02g
NaCl.....	0.02g
Uracil	0.02g
Pyridoxine·HCl	0.4mg
Riboflavin	0.4mg
p-Aminobenzoic acid.....	0.2mg
Calcium pantothenate	0.2mg
Niacin.....	0.2mg
Thiamine·HCl	0.2mg
Biotin	0.8μg

pH 6.7 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Add standard solutions and test solutions to each tube. Bring volume of each tube to 10.0mL. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the assay of tryptophan using *Lactobacillus plantarum* as an indicator organism.

Tryptophan Broth**Composition** per 100.0mL:

L-Tryptophan.....	0.5g
NaCl.....	0.5g
KH ₂ PO ₄	0.25g

pH 7.4 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to

100.0mL. Mix thoroughly. Adjust pH to 7.4. Filter sterilize. Aseptically distribute in 1.0mL volumes into sterile screw-capped tubes.

Use: Used to differentiate bacteria based on indole production. Indole is determined by the addition of modified Kovacs reagent to cultures that have incubated for 18–24 hr. Formation of a red color in the upper layer indicates indole formation.

Tryptose Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms.

Tryptose Agar (BAM M167)

Composition per liter:

Tryptose	20.0g
Agar	15.0g
NaCl	5.0g
Glucose	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes or leave in tubes. For slants allow tubes to cool in an inclined position.

Use: For the cultivation of a variety of bacteria for serology.

Tryptose Agar with Citrate

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
Sodium citrate	10.0g

NaCl	5.0g
Glucose	1.0g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms, including *Brucella* species and streptococci.

Tryptose Agar with Sheep Blood (ATCC Medium 546)

Composition per liter:

Agar	15.0g
Tryptose	10.0g
NaCl	5.0g
Beef extract	3.0g
Sheep blood, defibrinated	50-100mL
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components, except sheep blood to distilled/deionized water and bring volume to 900-950mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile sheep blood. Mix thoroughly. Pour into sterile Petri dishes in 17.0mL volumes or distribute into sterile tubes.

Use: For the cultivation and maintenance of a wide variety of fastidious microorganisms.

Tryptose Agar with Thiamine

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g
Thiamine·HCl	5.0mg
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of fastidious aerobic and facultative microorganisms, including *Brucella* species and streptococci.

Tryptose Blood Agar Base (BAM M166)

Composition per liter:

Agar	15.0g
Tryptose	10.0g
NaCl	5.0g
Beef extract	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C. Allow tubes to cool in a slanted position to obtain a 4–5.0cm slant and a 2–3.0cm butt.

Use: For the cultivation and enumeration of *Salmonella* species from foods.

Tryptose Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fastidious aerobic and facultative microorganisms, including streptococci.

Tryptose Broth

Composition per liter:

Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Glucose	1.0g
Thiamine-HCl	5.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fastidious aerobic and facultative microorganisms.

Tryptose Broth (BAM M167)

Composition per liter:

Tryptose	20.0g
NaCl	5.0g
Glucose	1.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of bacteria for serology.

Tryptose Broth with Citrate

Composition per liter:

Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
Sodium citrate	10.0g
NaCl	5.0g
Glucose	1.0g
Thiamine-HCl	5.0mg

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of a variety of fastidious aerobic microorganisms, especially *Brucella* species, from dairy products.

Tryptose Cycloserine Dextrose Agar

Composition per liter:

Agar	20.0g
Tryptose	15.0g
Pancreatic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
Cycloserine solution	10.0mL

pH 7.6 ± 0.2 at 25°C

Cycloserine Solution:

Composition per 10.0mL:

D-Cycloserine	0.4g

Preparation of Cycloserine Solution: Add cycloserine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloserine solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptical-

ly add sterile cycloserine solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of *Clostridium* species, especially *Clostridium botulinum*, from foods.

Tryptose Phosphate Broth

Composition per liter:

Tryptose	20.0g
NaCl	5.0g
Na ₂ HPO ₄	2.5g
Glucose	2.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Prior to the inoculation of anaerobic microorganisms, place tubes of sterile medium in a 100°C bath for 15 min and cool undisturbed.

Use: For the cultivation of a variety of fastidious bacteria.

Tryptose Phosphate Broth (TPB) (BAM M168)

Composition per liter:

Tryptose	20.0g
NaCl	5.0g
Na ₂ HPO ₄	2.5g
Glucose	2.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: For cell culture.

Tryptose Sulfite Cycloserine Agar (TSC Agar) (BAM M169)

Composition per liter:

Tryptose	15.0g
Agar	14.0g
Pancreatic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g

Na ₂ S ₂ O ₅	1.0g
Cycloserine solution	10.0mL
pH 7.6 ± 0.2 at 25°C	

Cycloserine Solution:

Composition per 10.0mL:

D-Cycloserine	0.4g

Preparation of Cycloserine Solution: Add cycloserine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloserine solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile cycloserine solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the presumptive identification and enumeration of *Clostridium perfringens*.

Tryptose Sulfite Cycloserine Agar (TSC Agar) (BAM M169)

Composition per liter:

Tryptose	15.0g
Agar	14.0g
Beef extract	5.0g
Pancreatic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
Na ₂ S ₂ O ₅	1.0g
Egg yolk emulsion	50.0mL
Cycloserine solution	10.0mL
pH 7.6 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Egg Yolk Emulsion:

Composition:

Chicken egg yolks	11
Whole chicken egg	1

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 1 chicken egg.

Cycloserine Solution:

Composition per 10.0mL:

D-Cycloserine	0.4g

Preparation of Cycloserine Solution: Add cycloserine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloserine solution and egg yolk emulsion, to

distilled/deionized water and bring volume to 940.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile cycloserine solution and egg yolk emulsion. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the presumptive identification and enumeration of *Clostridium perfringens*.

Tryptose Sulfite Cycloserine Agar with Polymyxin and Kanamycin

Composition per liter:

Tryptose	15.0g
Agar	14.0g
Beef extract	5.0g
Pancreatic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
Na ₂ S ₂ O ₅	1.0g
Antibiotic solution	10.0mL
pH 7.6 ± 0.2 at 25°C	

Antibiotic Solution:

Composition per 10.0mL:

D-Cycloserine	0.4g
Polymyxin B sulfate	0.03g
Kanamycin sulfate	0.012g

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except antibiotic solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile antibiotic solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the isolation and enumeration of *Clostridium perfringens* from foods and clinical specimens.

Tryptose Sulfite Cycloserine Agar without Egg Yolk (TSC Agar without Egg Yolk)

Composition per liter:

Tryptose	15.0g
Agar	14.0g
Beef extract	5.0g
Pancreatic digest of soybean meal	5.0g
Yeast extract	5.0g
Ferric ammonium citrate	1.0g
Na ₂ S ₂ O ₅	1.0g
Cycloserine solution	10.0mL
pH 7.6 ± 0.2 at 25°C	

Cycloserine Solution:

Composition per 10.0mL:

D-Cycloserine	0.4g

Preparation of Cycloserine Solution: Add cycloserine to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except cycloserine solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile cycloserine solution. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the presumptive identification and enumeration of *Clostridium perfringens*.

TS Agar (DSMZ Medium 893)

Composition per liter:

Agar	15.0g
Sucrose	5.0g
Tryptone	5.0g
Beef extract	3.0g
Glucose	1.0g
DpH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharococcus thermophilus*.

TSC Agar, Fluorocult (Fluorocult TSC Agar) (Fluorocult Tryptose Sulfite Cycloserine Agar) (Tryptose Sulfite Cycloserine Agar, Fluorocult)

Composition per liter:

Agar	15.0g
Tryptose	15.0g
Peptone from soymeal	5.0g
Yeast extract	5.0g
Na ₂ S ₂ O ₅	1.0g
Ammonium ferric citrate	1.0g
D-Cycloserine	0.2g
4-Methylumbelliferyl-phosphate disodium salt	50.0mg

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the isolation and enumeration of the vegetative and spore forms of *Clostridium perfringens* in foodstuffs. The culture medium complies with the recommendations of the International Organization for Standardization (ISO) (1978) and the DIN Norm 10165 for the examination of meat and meat products. It also conforms with the APHA recommendations for the examination of foods (1992). D-Cycloserine inhibits the accompanying bacterial flora and causes the colonies which develop to remain smaller. 4-Methylumbelliferyl-phosphate (MUP) is a fluorogenic substrate for the alkaline and acid phosphatase. The acid phosphatase is a highly specific indicator for *C. perfringens*. The acid phosphatase splits the fluorogenic substrate MUP forming 4-methylumbelliferone which can be identified by its fluorescence in long wave UV light. Thus a strong suggestion for the presence of *C. perfringens* can be obtained.

TSN Agar

(Trypticase Sulfite Neomycin Agar)

Composition per liter:

Pancreatic digest of casein	15.0g
Agar	13.5g
Yeast extract	10.0g
Na ₂ SO ₃	1.0g
Ferric citrate	0.5g
Neomycin sulfate	0.05g
Polymyxin sulfate	0.02g
Buffered thioglycolate solution	50.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Buffered Thioglycolate Solution:

Composition per 50.0mL:

Buffer solution	35.0mL
Sodium thioglycolate solution	15.0mL

Preparation of Buffered Thioglycolate Solution: Combine components. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C.

Buffer Solution:

Composition per 100.0mL:

Na ₂ CO ₃	28.0g
K ₂ HPO ₄	5.7g

Preparation of Buffer Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Thioglycolate Solution:

Composition per 100.0mL:

Sodium thioglycolate	13.3g

Preparation of Thioglycolate Solution: Add sodium thioglycolate to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except buffered thioglycolate solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 12 min at 13 psi pressure–118°C. Do not overheat. Cool to 45°–50°C. Aseptically add buffered thioglycolate solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the selective isolation of *Clostridium perfringens*.

TSY Medium

(Trypticase Soy

Yeast Extract Medium)

Agar	20.0g
Pancreatic digest of casein	17.0g
Yeast extract	5.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

TSYES Medium

(Trypticase Soy Yeast Extract Starch Medium)

Composition per liter:

Pancreatic digest of casein	17.0g
Agar	15.0g
NaCl	5.0g
Papaic digest of soybean meal	3.0g
K ₂ HPO ₄	2.5g
Glucose	2.5g

Yeast extract.....	2.0g
Soluble starch.....	1.0g
pH 7.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Bacillus* species.

Tween 80 Hydrolysis Broth

Composition per liter:

Na ₂ HPO ₄	5.79g
NaH ₂ PO ₄	3.53g
Neutral Red.....	0.02g
Tween 80.....	5.0mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of *Mycobacterium* species. Strains that hydrolyze Tween 80 within 5 days turn the medium pink to red.

Tween 80 Hydrolysis Broth

Composition per 125.0mL:

Neutral Red.....	0.1g
Solution 1.....	38.9mL
Solution 2.....	61.1mL
Tween 80.....	25.0mL
pH 7.0 ± 0.2 at 25°C	

Solution 1:

Composition per 400.0mL:

KH ₂ PO ₄	22.7g

Preparation of Solution 1: Add KH₂PO₄ to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly.

Solution 2:

Composition per 400.0mL:

Na ₂ HPO ₄	23.8g

Preparation of Solution 2: Add Na₂HPO₄ to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of *Mycobacterium* species. Strains that hydrolyze Tween 80 within 5 days turn the medium pink to red.

Tween 80 Hydrolysis Broth

Composition per 102.5mL:

NaHPO ₄ (0.066M solution).....	61.1mL
KH ₂ PO ₄ (0.066M solution).....	38.9mL
Neutral Red (0.1% solution).....	2.0mL
Tween 80.....	0.5mL
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Combine components. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the differentiation of *Mycobacterium* species. Strains that hydrolyze Tween 80 within 5 days turn the medium pink to red.

Tween 80 Hydrolysis Medium

Composition per liter:

Agar.....	12.0g
Peptone.....	10.0g
NaCl.....	5.0g
CaCl ₂	0.1g
Tween 80.....	10.0mL
pH 7.2–7.4 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes.

Use: For the cultivation and differentiation of *Pseudomonas* species based on their ability to hydrolyze Tween 80. Bacteria that hydrolyze Tween 80 appear as colonies surrounded by an opaque zone.

TY Medium

Composition per liter:

Agar.....	15.0g
Pancreatic digest of casein.....	5.0g
Yeast extract.....	3.0g
CaCl ₂ ·6H ₂ O.....	1.3g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a wide variety of bacteria.

TY Medium, 2X**Composition per liter:**

Pancreatic digest of casein.....	16.0g
Yeast extract.....	10.0g
NaCl.....	5.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli*.

TY Salt Medium**Composition per liter:**

NaCl.....	10.0g
Pancreatic digest of casein.....	10.0g
Yeast extract.....	5.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of bacteria.

TYES Medium**(Tryptone Yeast Extract Salt Medium)****Composition per liter:**

Agar.....	15.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g
Yeast extract.....	1.0g
CaCl ₂	0.3g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Escherichia coli*.

TYG Medium**(Trypticase Yeast Extract****Glucose Medium)****(ATCC Medium 603)****Composition per liter:**

Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g
Yeast extract.....	1.0g

Glucose.....	1.0g
CaCl ₂ ·2H ₂ O.....	0.3g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of *Escherichia coli*.

TYGPN Medium**Composition per liter:**

Pancreatic digest of casein.....	20.0g
KNO ₃	10.0g
Yeast extract.....	10.0g
Na ₂ HPO ₄	5.0g
Glycerol (80% solution).....	10.0mL

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 25 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli*.

TYGS Medium**(Tryptone Yeast Extract
Glucose Salt Medium)****Composition per liter:**

Agar.....	15.0g
Pancreatic digest of casein.....	10.0g
NaCl.....	8.0g
Yeast extract.....	1.0g
CaCl ₂ ·2H ₂ O solution.....	100.0mL
Glucose solution.....	100.0mL

CaCl₂·2H₂O Solution:**Composition per 100.0mL:**

CaCl ₂ ·2H ₂ O.....	0.3g

Preparation of CaCl₂·2H₂O Solution: Add the CaCl₂·2H₂O to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Glucose Solution:**Composition per 100.0mL:**

D-Glucose.....	1.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except CaCl₂·2H₂O solution and glucose solution, to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add the sterile

CaCl₂·2H₂O solution and sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of a variety of bacteria.

Tyrosine Agar (BAM M170)

Composition per 1100mL:

Base solution.....	1.0L
Tyrosine suspension.....	100.0mL
pH 7.0 ± 0.2 at 25°C	

Base Solution:

Composition per 1.0L:

Agar	15.0g
Pancreatic digest of gelatin	5.0g
Beef extract	3.0g

Preparation of Base Solution: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48°C.

Tyrosine Suspension:

Composition per 100.0mL:

Tyrosine	5.0g

Preparation of Tyrosine Suspension: Add tyrosine to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 48°C.

Preparation of Medium: Combine 1.0L sterile base solution with 100.0mL sterile tyrosine suspension. Mix thoroughly by swirling or inverting several times. Distribute into sterile tubes or pour into sterile Petri dishes. Cool rapidly to prevent tyrosine from separating. For slants allow tubes to cool in an inclined position.

Use: For the selective isolation of *Brevibacillus brevis* and *Bacillus cereus*.

TYX Medium

Composition per liter:

Yeast extract.....	5.0g
K ₂ HPO ₄	3.5g
Tryptone.....	1.0g
Na ₂ SO ₄	0.2g
L-cysteinium chloride·H ₂ O.....	0.5g
Biotin	10.0mg
<i>p</i> -Aminobenzoic acid.....	10.0mg
Sugar solution	5.0mL
Resazurin	4.0mL
Trace elements solution	1.0mL

Resazurin Solution:

Composition per 100.0mL:

Resazurin	25mg

Preparation of Resazurin Solution: Add resazurin to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Trace Elements Solution:

Composition per 100.0mL:

FeCl ₃ ·6H ₂ O.....	2.7g
MgSO ₄	1.2g
NaMoO ₄ ·2H ₂ O	0.24g
MnSO ₄ ·7H ₂ O.....	0.17g
CaCl ₂ ·2H ₂ O	0.15g
ZnSO ₄ ·7H ₂ O.....	29.0mg
CuSO ₄ ·5H ₂ O.....	25.0mg
CoCl ₂ ·6H ₂ O.....	24.0mg
H ₂ SO ₄	2.8mL

Preparation of Trace Elements Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Sugar Solution:

Composition per 100.0mL:

Xylose.....	1.0g

Preparation of Resazurin Solution: Add xylose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteinium chloride, biotin, *p*-aminobenzoic acid, and sugar solution, to distilled/deionized water and bring volume to 995.0mL. Mix thoroughly. Adjust pH to 7.0. Gently heat but do not boil. Cook for 5-10 min so that color first turns red and then turns yellow. Cool on ice. Add L-cysteinium chloride, biotin, and *p*-aminobenzoic acid. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 5.0mL of sterile sugar solution. Mix thoroughly.

Use: For the cultivation of *Clostridium* species.

UBA Medium (Universal Beer Agar)

Composition per liter:

Glucose	16.1g
Peptonized milk	15.0g
Agar	12.0g
Tomato juice, dessicated.....	12.2g
Yeast extract.....	6.1g
K ₂ HPO ₄	0.31g
KH ₂ PO ₄	0.31g
MgSO ₄ ·7H ₂ O	0.12g
FeSO ₄	6.0mg
MnSO ₄ ·5H ₂ O	6.0mg

NaCl.....	6.0mg
Beer.....	250.0mL
pH 6.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components, except beer, to distilled/deionized water and bring volume to 750.0mL. Mix thoroughly. Gently heat and bring to boiling. Add beer. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For cultivation of microorganisms of significance in the brewing industry.

Universal Beer Agar

Composition per liter:

Peptonized milk	15.0g
Agar	12.0g
Yeast extract.....	10.0g
Glucose	10.0g
Tomato juice solids	7.0g
K ₂ HPO ₄	0.5g
KH ₂ PO ₄	0.5g
MgSO ₄ ·5H ₂ O	0.2g
NaCl.....	0.01g
FeSO ₄ ·7H ₂ O.....	0.01g
MnSO ₄ ·H ₂ O	0.01g
Beer.....	250.0mL
pH 6.3 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components, except beer, to distilled/deionized water and bring volume to 750.0mL. Mix thoroughly. Gently heat and bring to boiling. Add beer. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 10 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 250.0mL of beer without degassing. Pour into sterile Petri dishes or leave in tubes.

Use: For the enumeration of contaminating bacteria and yeasts encountered in wort and beer.

Universal Medium for Yeasts (YM) (DSMZ Medium 186)

Composition per liter:

Agar	15.0g
Glucose	10.0g
Peptone.....	5.0g
Yeast extract.....	3.0g
Malt extract.....	3.0g
pH 6.8 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces* spp. and other yeasts.

Universal Preenrichment Broth (BAM M188)

Composition per liter:

KH ₂ PO ₄	15.0g
Na ₂ HPO ₄	7.0g
Tryptone.....	5.0g
Proteose peptone.....	5.0g
NaCl.....	5.0g
Glucose	0.5g
MgSO ₄	0.25g
Sodium pyruvate.....	0.2g
Ferric ammonium citrate.....	0.1g
pH 6.3 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat until dissolved. Adjust pH to 6.3. Distribute into tubes in 10.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the enrichment of injured foodborne pathogens of different genera simultaneously in lieu of having to undergo separate simultaneous enrichment cultures for subsequent detection or isolation of each pathogen.

Urea Agar

Composition per liter:

Agar	15.0g
Urea.....	10.0g
Na ₂ HPO ₄ ·12H ₂ O	9.0g
NaCl.....	5.0g
KH ₂ PO ₄	1.5g
Meat extract	1.0g
Yeast extract.....	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
MnCl ₂ ·4H ₂ O	20.0mg
CaCl ₂	1.2mg
Glucose solution	100.0mL

Glucose Solution:

Composition per 100.0mL:

Glucose	5.0g

Preparation of Glucose Solution: Add glucose to distilled/deionized water and bring volume to

100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except glucose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile glucose solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Corynebacterium glutamicum*.

Urea Agar

Composition per liter:

Agar	15.0g
Na ₂ HPO ₄ ·12H ₂ O	9.0g
NaCl	5.0g
KH ₂ PO ₄	1.5g
Meat extract	1.0g
Yeast extract	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnCl ₂ ·4H ₂ O	20.0mg
CaCl ₂	1.2mg
Glucose-urea solution	100.0mL

Glucose-Urea Solution:

Composition per 100.0mL:

Urea	10.0g
Glucose	5.0g

Preparation of Glucose-Urea Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except glucose-urea solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile glucose-urea solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Corynebacterium glutamicum*.

Urea Agar

(Urease Test Agar)

(Urea Agar Base, Christensen)

Composition per liter:

Urea	20.0g
Agar	15.0g
NaCl	5.0g
KH ₂ PO ₄	2.0g

Peptone	1.0g
Glucose	1.0g
Phenol Red	0.012g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components, except agar, to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Add agar to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add the 100.0mL of sterile basal medium. Mix thoroughly. Distribute into sterile tubes. Allow tubes to solidify in a slanted position.

Use: For the differentiation of a variety of microorganisms, especially members of the *Enterobacteriaceae*, aerobic actinomycetes, streptococci, and nonfermenting Gram-negative bacteria, on the basis of urease production.

Urea Broth (Urease Test Broth) (BAM M171)

Composition per liter:

Urea	20.0g
Na ₂ HPO ₄	9.5g
KH ₂ PO ₄	9.1g
Yeast extract	0.1g
Phenol Red	0.01g
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostics.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Do not heat. Filter sterilize. Aseptically distribute into sterile tubes or flasks.

Use: For the differentiation of organisms, especially the *Enterobacteriaceae*, on the basis of urease production. Urease-positive bacteria turn the medium pink.

Urea Broth Base

Composition per liter:

NaCl	5.0g
Na ₂ HPO ₄	1.2g
Peptone	1.0g
Glucose	1.0g
KH ₂ PO ₄	0.8g
Phenol Red	0.012g
Urea solution	50.0mL
pH 6.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Urea Solution:

Composition per 100.0mL:

Urea.....40.0g

Preparation of Urea Solution: Add urea to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except urea solution, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Autoclave for 20 min at 10 psi pressure–115°C. Cool to 50°C. Aseptically add 50.0mL of sterile urea solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the differentiation of members of the *Enterobacteriaceae* based on urease production. Urease-positive bacteria turn the medium pink.

Urea R Broth (Urea Rapid Broth) (BAM M172)

Composition per liter:

Urea.....20.0g
Yeast extract.....0.1g
Na₂HPO₄.....0.095g
KH₂PO₄.....0.091g
Phenol Red.....0.01g

pH 6.9 ± 0.2 at 25°C

Source: This medium is available as a prepared medium from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Aseptically distribute into sterile tubes or flasks.

Use: For the differentiation of members of the *Enterobacteriaceae* based on the rapid detection of urease activity. Urease-positive bacteria turn the medium cerise.

Urea Semisolid Medium

Composition per liter:

Solution A.....400.0mL
Solution B.....50.0mL

Solution A:

Composition per 400.0mL:

Pancreatic digest of casein.....6.0g
Yeast extract.....2.0g
NaCl.....1.0g
Yeast extract.....0.8g
Agar.....0.3g

L-Cystine.....0.1g
Thioglycollic acid.....0.12mL
pH 7.2 ± 0.2 at 25°C

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 400.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 60°C.

Solution B:

Composition per 50.0mL:

Urea.....8.0g
Na₂HPO₄.....3.8g
KH₂PO₄.....3.64g
Yeast extract.....0.04g
Phenol Red.....4.0mg

Preparation of Solution B: Add components to distilled/deionized water and bring volume to 50.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine 400.0mL of sterile solution A and 50.0mL of sterile solution B. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes in 7.0mL volumes. Pass the tubes into an anaerobic chamber containing 85% N₂ + 10% H₂ + 5% CO₂ for 60 min. Close screw caps tightly.

Use: For the cultivation and differentiation of anaerobic bacteria based on their production of urease. Bacteria that produce urease turn the medium bright red.

Urea Test Broth

Composition per liter:

Urea.....20.0g
Na₂HPO₄.....9.5g
KH₂PO₄.....9.1g
Yeast extract.....0.1g
Phenol Red.....0.01g
Urea solution.....100.0mL

Urea Solution:

Composition per 100.0mL:

Urea.....20.0g

Preparation of Urea Solution: Add urea to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except urea solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile urea solution. Mix thoroughly. Aseptically distribute into sterile tubes in 3.0mL volumes.

Use: For the cultivation and differentiation of members of the *Enterobacteriaceae* and aerobic actinomycetes based on their production of urease. Bacteria that produce urease turn the medium bright red.

Urease Test Broth (Urea Broth)

Composition per liter:

Urea.....	20.0g
Na ₂ HPO ₄	9.5g
KH ₂ PO ₄	9.1g
Yeast extract.....	0.1g
Phenol Red.....	0.01g

pH 6.8 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize. Aseptically distribute into sterile tubes or flasks.

Use: For the differentiation of organisms, especially the *Enterobacteriaceae*, on the basis of urease production. Urease-positive bacteria turn the medium pink.

Uric Acid Agar

Composition per liter:

Agar.....	15.0g
Na ₂ HPO ₄ ·12H ₂ O.....	9.0g
NaCl.....	5.0g
KH ₂ PO ₄	1.5g
Meat extract.....	1.0g
Yeast extract.....	1.0g
MgSO ₄ ·7H ₂ O.....	0.2g
MnCl ₂ ·4H ₂ O.....	20.0mg
CaCl ₂	1.2mg
Glucose-uric acid solution.....	100.0mL

Glucose-Uric Acid Solution:

Composition per 100.0mL:

Glucose.....	5.0g
Uric acid.....	0.4g

Preparation of Glucose-Uric Acid Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Warm to 50°C.

Preparation of Medium: Add components, except glucose-uric acid solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile glucose-uric acid solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Bacillus* species.

Uric Acid Agar for Clostridia

Composition per liter:

Agar.....	20.0g
K ₂ HPO ₄	4.0g
Uric acid.....	3.0g
Yeast extract.....	1.0g
Sodium thioglycolate.....	0.5g
MgSO ₄ ·7H ₂ O.....	0.1g
FeSO ₄ ·7H ₂ O.....	5.0mg
Phenol Red (0.04% solution).....	1.0mL

pH 7.6–8.0 at 25°C

Preparation of Medium: Add components, except uric acid, to approximately 900.0mL of distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.6 with 1N NaOH. Add the uric acid. Mix thoroughly. Adjust pH to 7.6. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of anaerobic bacteria, such as *Clostridium acidurici* and *Clostridium cylindrosporium*, that can utilize uric acid as sole source of carbon and energy.

Uric Acid Semisolid Agar for Clostridia

Composition per liter:

Agar.....	2.0g
K ₂ HPO ₄	4.0g
Uric acid.....	3.0g
Yeast extract.....	1.0g
Sodium thioglycolate.....	0.5g
MgSO ₄ ·7H ₂ O.....	0.1g
FeSO ₄ ·7H ₂ O.....	5.0mg
Phenol Red (0.04% solution).....	1.0mL

pH 7.6–8.0 at 25°C

Preparation of Medium: Add components, except uric acid, to approximately 900.0mL of distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.6 with 1N NaOH. Add the uric acid. Mix thoroughly. Adjust pH to 7.6. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and maintenance of anaerobic bacteria, such as *Clostridium acidurici* and *Clostridium cylindrosporium*, that can utilize uric acid as sole source of carbon and energy.

Uric Acid Broth

Composition per liter:

Na ₂ HPO ₄ ·12H ₂ O.....	9.0g
NaCl.....	5.0g

KH ₂ PO ₄	1.5g
Meat extract	1.0g
Yeast extract	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnCl ₂ ·4H ₂ O	20.0mg
CaCl ₂	1.2mg
Glucose-uric acid solution	100.0mL

Glucose-Uric Acid Solution:**Composition per 100.0mL:**

Glucose	5.0g
Uric acid	0.4g

Preparation of Glucose-Uric Acid Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium:

Add components, except glucose-uric acid solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 100.0mL of sterile glucose-uric acid solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation and maintenance of *Bacillus* species.

UVM *Listeria* Enrichment Broth**(University of Vermont *Listeria* Enrichment Broth)****Composition per liter:**

NaCl	20.0g
Na ₂ HPO ₄	9.6g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	1.35g
Esculin	1.0g
Nalidixic acid	40.0mg
Acriflavine·HCl	12.0mg

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation of *Listeria monocytogenes*.

UVM Modified***Listeria* Enrichment Broth****(University of Vermont Modified*****Listeria* Enrichment Broth)****Composition per liter:**

NaCl	20.0g
Na ₂ HPO ₄	9.6g
Pancreatic digest of casein	5.0g
Peptic digest of animal tissue	5.0g
Beef extract	5.0g
Yeast extract	5.0g
KH ₂ PO ₄	1.35g
Esculin	1.0g
Nalidixic acid	20.0mg
Acriflavine·HCl	12.0mg

pH 7.2 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the selective isolation of *Listeria monocytogenes*.

V-8™ Agar**Composition per liter:**

Agar	15.0g
CaCO ₃	2.0g
V-8 canned vegetable juice	200.0mL

Preparation of Medium:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of numerous yeasts and filamentous fungi.

V-8 Juice Agar II**(ATCC Medium 2040)****Composition per liter:**

Agar	30.0g
V-8 canned vegetable juice	300.0mL

pH 7.3 ± 0.2 at 25°C

Preparation of Medium:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of various yeasts and filamentous fungi.

Veal Infusion Agar (BAM M173)

Composition per liter:

Veal, infusion from	500.0g
Agar	15.0g
Proteose peptone No.3	10.0g
NaCl	5.0g
pH 7.3 ± 0.2 at 25°C	

Source: This medium is available from BD Diagnostics.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Pour into sterile Petri dishes or leave in tubes. For slants allow tubes to cool in an inclined position.

Use: For the cultivation of fastidious microorganisms.

Veal Infusion Broth (BAM M173)

Composition per liter:

Veal, infusion from	500.0g
Proteose peptone No.3	10.0g
NaCl	5.0g
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available from BD Diagnostics.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of fastidious microorganisms.

***Vibrio* Agar**

Composition per liter:

Sucrose	20.0g
Agar	15.0g
NaCl	10.0g
Sodium citrate·2H ₂ O	10.0g
Na ₂ S ₂ O ₃ ·5H ₂ O	6.5g
Oxgall	5.0g
Yeast extract	5.0g
Pancreatic digest of casein	4.0g
Proteose peptone	3.0g
Sodium deoxycholate	1.0g

Sodium lauryl sulfate	0.2g
Water Blue	0.2g
Cresol Red	0.02g
pH 8.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.5. Gently heat and bring to boiling. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the isolation and cultivation of the *Vibrio cholerae*.

***Vibrio parahaemolyticus* Agar (VP Agar)**

Composition per liter:

Agar	20.0g
NaCl	20.0g
Sucrose	20.0g
Sodium citrate	10.0g
Na ₂ S ₂ O ₃ ·5H ₂ O	10.0g
Peptone	10.0g
Sodium taurocholate	5.0g
Yeast extract	5.0g
Sodium lauryl sulfate	0.2g
Bromthymol Blue	0.04g
Thymol Blue	0.04g
pH 8.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Pour into sterile Petri dishes.

Use: For the isolation, cultivation, enumeration, and presumptive identification of coliforms in milk, food, and other specimens of sanitary significance. For the enumeration of bacteria in cheese, especially *Pseudomonas fragi*, *Pseudomonas viscosa*, and *Alcaligenes metalcaligenes*. Sucrose-fermenting bacteria appear as yellow colonies with pale yellow peripheries. Sucrose-nonfermenting bacteria appear as mucoid, green colonies with a dark green center.

***Vibrio parahaemolyticus* Sucrose Agar (VPSA) (BAM M191)**

Composition per liter:

NaCl	30.0g
Agar	15.0g
Sucrose	10.0g
Yeast extract	7.0g
Tryptose	5.0g

Pancreatic digest of casein.....	5.0g
Bile salts No. 3.....	1.5g
Bromthymol Blue	0.025g
pH 8.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Cool to 50°C. Pour into sterile Petri dishes in 20.0mL volumes. Allow plates to dry before using.

Use: For the isolation, cultivation, and differentiation of *Vibrio parahaemolyticus* from seafood. *Vibrio parahaemolyticus* and *Vibrio vulnificus* appear as blue to green colonies. Other *Vibrio* species appear as yellow colonies.

***Vibrio vulnificus* Agar (VVA) (BAM M190)**

Composition per liter:

NaCl.....	30.0g
Agar	25.0g
Peptone.....	20.0g
Cellobiose solution	100.0mL
Dye solution.....	10.0mL
pH 8.2 ± 0.2 at 25°C	

Dye Solution:

Composition per 100.0mL:

Bromthymol Blue	0.6g
Ethanol, 70%.....	100.0mL

Preparation of Dye Solution: Add Bromthymol Blue to 100.0mL of 70% ethanol. Mix thoroughly.

Cellobiose Solution:

Composition per 100.0mL:

Cellobiose	10.0g

Preparation of Cellobiose Solution: Add cellobiose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Gently heat while mixing to dissolve the cellobiose. Cool. Filter sterilize.

Preparation of Medium: Add components, except cellobiose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat until dissolved. Adjust pH to 8.2. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 100.0mL sterile cellobiose solution. Mix thoroughly and pour into sterile Petri dishes. Final color of medium should be light blue.

Use: For the detection of *Vibrio vulnificus* from seafoods.

Violet Peptone Bile Lactose Broth

Composition per liter:

Lactose.....	10.0g
Peptone	10.0g
Bile salts.....	5.0g
Gentian Violet.....	0.04g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the selective cultivation of members of the *Enterobacteriaceae*.

Violet Red Bile Agar

Composition per liter:

Agar	15.0g
Lactose.....	10.0g
Glucose	10.0g
Pancreatic digest of gelatin.....	7.0g
NaCl.....	5.0g
Yeast extract.....	3.0g
Bile salts.....	1.5g
Neutral Red.....	0.03g
Crystal Violet.....	2.0mg
pH 7.4 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour immediately into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of members of the *Enterobacteriaceae* from brined vegetables. For the enumeration of members of the *Enterobacteriaceae* from brined vegetables by the pour plate technique.

Violet Red Bile Agar (VRB Agar) (BAM M174)

Composition per liter:

Agar	15.0g
Lactose.....	10.0g
Pancreatic digest of gelatin.....	7.0g
NaCl.....	5.0g
Yeast extract.....	3.0g
Bile salts.....	1.5g

Neutral Red	0.03g
Crystal Violet	2.0mg
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour immediately into sterile Petri dishes or leave in tubes.

Use: For the detection of coliform bacteria in food.

Violet Red Bile Agar with MUG (VRBA-MUG) (BAM M175)

Composition per liter:

Agar	15.0g
Lactose	10.0g
Pancreatic digest of gelatin	7.0g
NaCl	5.0g
Yeast extract	3.0g
Bile salts	1.5g
MUG (4-methylumbelliferyl- β-D-glucuronide)	0.1g
Neutral Red	0.03g
Crystal Violet	2.0mg
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour immediately into sterile Petri dishes or leave in tubes.

Use: For the differentiation of *Escherichia coli* from dairy products and other foods based on their ability to produce β-glucuronidase.

Violet Red Bile Glucose Agar

Composition per liter:

Agar	12.0g
Glucose	10.0g
Peptone	7.0g
NaCl	5.0g
Yeast extract	3.0g
Bile salts No. 3	1.5g

Neutral Red	0.03g
Crystal Violet	2.0mg
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not autoclave. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the detection and enumeration of *Enterobacteriaceae* from foods.

Vitamin B₁₂ Assay Medium

Composition per liter:

Glucose	40.0g
Sodium acetate	20.0g
Vitamin assay casamino acids	12.0g
Sorbitan monooleate complex	2.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
DL-Tryptophan	0.2g
L-Cystine	0.2g
Adenine	0.02g
FeSO ₄	0.02g
Guanine	0.02g
MnSO ₄ ·5H ₂ O	0.02g
NaCl	0.02g
Uracil	0.02g
Pyridoxine·HCl	4.0mg
Niacin	2.0mg
Riboflavin	2.0mg
Thiamine·HCl	2.0mg
Xanthine	1.0mg
Calcium pantothenate	200μg
p-Aminobenzoic acid	200μg
Folic acid	100μg
Biotin	10μg

pH 6.3 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 5.0mL volumes. Add standard solution or test solutions to each tube. Adjust the volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the microbiological assaying of vitamin B₁₂ using *Lactobacillus leichmannii* as the test organism.

Vitamin B₁₂ Assay Medium with Colistin

Composition per liter:

Glucose	40.0g
Sodium acetate.....	20.0g
Vitamin assay casamino acids.....	12.0g
Sorbitan monooleate complex	2.0g
K ₂ HPO ₄	1.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.4g
Colistin sulfate	0.5g
DL-Tryptophan.....	0.2g
L-Cystine	0.2g
Adenine.....	0.02g
FeSO ₄	0.02g
Guanine.....	0.02g
MnSO ₄ ·5H ₂ O	0.02g
NaCl.....	0.02g
Uracil	0.02g
Pyridoxine-HCl	4.0mg
Niacin.....	2.0mg
Riboflavin	2.0mg
Thiamine-HCl	2.0mg
Xanthine.....	1.0mg
Calcium DL-pantothenate.....	200µg
<i>p</i> -Aminobenzoic acid.....	200µg
Folic acid.....	100µg
Biotin	10µg
Cyanocobalamin	250.0ng

pH 6.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Continue boiling for 2–3 min. Distribute into tubes in 5.0mL volumes. Add standard solution or test solutions to each tube. Adjust the volume of each tube to 10.0mL with distilled/deionized water. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus leichmannii*.

Vogel and Johnson Agar (VJ Agar) (BAM M176)

Composition per liter:

Agar	15.0g
Pancreatic digest of casein.....	10.0g
D-Mannitol	10.0g
Glycine.....	10.0g

Yeast extract.....	5.0g
K ₂ HPO ₄	5.0g
LiCl	5.0g
Phenol Red.....	0.025g
K ₂ TeO ₃ solution	20.0mL
pH 7.2 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

K₂TeO₃ Solution:

Composition per 100.0mL:

K ₂ TeO ₃	1.0g

Preparation of K₂TeO₃ Solution: Add K₂TeO₃ to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Caution: Potassium tellurite is toxic.

Preparation of Medium: Add components, except K₂TeO₃ solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 20.0mL of sterile K₂TeO₃ solution. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the detection of coagulase-positive *Staphylococcus aureus*.

Voges Proskauer Medium (VP Broth, Modified) (BAM M177)

Composition per liter:

Proteose peptone.....	7.0g
Glucose	5.0g
NaCl.....	5.0g

pH 6.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes in 5.0mL volumes. Autoclave for 10 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Bacillus cereus* from foods.

VP Medium (Voges-Proskauer Medium)

Composition per liter:

Peptone.....	7.0g
K ₂ HPO ₄	5.0g
Glucose	5.0g

pH 6.9 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.9. Distribute into tubes in 3.0mL volumes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and differentiation of bacteria based on their ability to produce acetoin.

VRB Agar, Fluorocult Fluorocult VRB Agar

Composition per liter:

Agar	13.0g
Lactose	10.0g
Peptone from meat	7.0g
NaCl	5.0g
Yeast extract	3.0g
Bile salt mixture	1.5g
4-Methylumbelliferyl- β -D-glucuronide	0.1g
Neutral Red	0.03g
Crystal Violet	0.002g

pH 7.4 $\pm$ 0.2 at 25°C

Source: This medium is available from Merck.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Heat in a boiling water bath or in free flowing steam with frequent stirring until completely dissolved. Do not boil for more than 2 min. Do not autoclave. Do not overheat. Pour into sterile Petri dishes. The plates are clear and dark red.

Use: For the detection and enumeration of coliform bacteria, in particular *E. coli*. Crystal violet and bile salts largely inhibit the growth of Gram-positive accompanying bacterial flora. Lactose-positive colonies show a color change to red of the pH indicator. *E. coli* colonies show a fluorescence under UV light. Lactose-negative *Enterobacteriaceae* are colorless. Lactose-positive colonies are red and often surrounded by a turbid zone due to the precipitation of bile acids. Light blue fluorescing colonies denote *E. coli*.

Wagatsuma Agar (BAM M178)

Composition per 1050.0mL:

NaCl	70.0g
Agar	15.0g
Mannitol	10.0g
Peptone	10.0g
K ₂ HPO ₄	5.0g
Yeast extract	3.0g
Crystal Violet	1.0mg
Red blood cells	50.0mL

pH 8.0 $\pm$ 0.2 at 25°C

Red Blood Cells:

Composition per 100.0mL:

Blood, human or rabbit	100.0mL

Preparation of Red Blood Cells: Mix freshly drawn human or rabbit blood with anticoagulant and an equal volume of sterile 0.85% saline solution. Centrifuge cells at 4000 $\times$ g at 4°C for 15 min. Pour off saline and wash two more times with sterile saline. After last wash, pour off saline and resuspend cells to their original volume.

Preparation of Medium: Add components, except blood, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 8.0. Place in a steam bath for 30 min. Do not autoclave. Cool to 45°–50°C. Add 50.0mL of washed red blood cells. Mix thoroughly. Pour into sterile Petri dishes. Dry plates before using.

Use: For the cultivation and detection of thermally stable hemolysis of *Vibrio parahaemolyticus* by the Kanagawa reaction.

Wallenstein Medium

Composition per 4.225L:

Malachite Green	0.75g
Egg yolk emulsion	3.125L
Glycerol	100.0mL

pH 6.75 $\pm$ 0.2 at 25°C

Egg Yolk Emulsion:

Composition:

Chicken egg yolks	66
Whole chicken egg	6

Preparation of Egg Yolk Emulsion: Soak eggs with 1:100 dilution of saturated mercuric chloride solution for 1 min. Crack eggs and separate yolks from whites. Mix egg yolks with 6 chicken eggs.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation of *Mycobacterium* species.

Wang's Transport Storage Medium

Composition per liter:

Pancreatic digest of casein	10.0g
Peptic digest of animal tissue	10.0g
NaCl	5.0g
Agar	4.0g
Yeast extract	2.0g
Glucose	1.0g
NaHSO ₃	0.1g
Sheep blood, defibrinated	100.0mL

pH 7.0 $\pm$ 0.2 at 25°C

Preparation of Medium: Add components, except sheep blood, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add sterile sheep blood. Sheep blood may be replaced by 50.0mL of horse blood and 50.0mL of sterile distilled/deionized water. Mix thoroughly. Aseptically distribute into sterile screw-capped tubes in 4.0mL volumes. Allow tubes to cool in an upright position.

Use: For the cultivation, transport, and maintenance of *Campylobacter* species from foods.

Waxy Maize Starch Medium

Composition per liter:

Agar	20.0g
Waxy maize starch	5.0g
Pancreatic digest of casein	5.0g
Yeast extract	5.0g
CoCl ₂ ·6H ₂ O	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Maltose solution	100.0mL
pH 6.7 ± 0.2 at 25°C	

Maltose Solution:

Composition per 100.0mL:

Maltose	10.0g

Preparation of Maltose Solution: Add maltose to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except maltose solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 6.7. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add maltose solution. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* species.

Wesley Broth

Composition per liter:

Tryptose	20.0g
Bicine (N,N-bis-2-[hydroxy-ethyl]glycine) buffer	10.0g
NaCl	5.0g
Yeast extract	2.5g
Agar	1.0g
FeSO ₄	0.25g
Na ₂ S ₂ O ₅	0.25g
Sodium pyruvate	0.25g
Antibiotic solution	10.0mL
Alkaline hematin solution	6.25mL

Antibiotic Solution:

Composition per 10.0mL:

Rifampin	0.025g
Cefsulodin	6.25mg
Polymyxin B sulfate	20,000U

Preparation of Antibiotic Solution: Add components to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Alkaline Hematin Solution:

Composition per 10.0mL:

Hemin	0.032g
NaOH (0.15N solution)	10.0mL

Preparation of Alkaline Hematin Solution:

Add hemin to 10.0mL of NaOH solution. Mix thoroughly. Autoclave for 30 min at 5 psi pressure–108°C. Cool to 25°C.

Preparation of Medium: Add components, except antibiotic solution and alkaline hematin solution, to distilled/deionized water and bring volume to 983.75mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 10.0mL of sterile antibiotic solution and 6.25mL of sterile alkaline hematin solution. Mix thoroughly. Aseptically distribute into sterile tubes. Use medium immediately or store overnight at 4°C.

Use: For the enrichment of *Campylobacter* species from foods.

Whey Agar

Composition per liter:

Whey permeate	50.0g
Casein hydrolysate	20.0g
Agar	15.0g
Yeast extract	10.0g
Tween 80	1.0mL
pH 5.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.6. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus* species.

Whey Agar

Composition per liter:

Whey permeate	50.0g
Casein hydrolysate	20.0g
Agar	15.0g

Yeast extract.....	10.0g
Tween 80.....	1.0mL
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactococcus* species.

Whey Broth

Composition per liter:

Whey permeate.....	50.0g
Casein hydrolysate.....	20.0g
Yeast extract.....	10.0g
Tween 80.....	1.0mL
pH 5.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 5.6. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus* species.

Whey Broth

Composition per liter:

Whey permeate.....	50.0g
Casein hydrolysate.....	20.0g
Yeast extract.....	10.0g
Tween 80.....	1.0mL
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.5. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactococcus* species.

Wickerham Broth

Composition per 100.0mL:

Carbohydrate.....	10.0g
Yeast nitrogen base.....	100.0mL

Yeast Nitrogen Base, 10X:

Composition per liter:

Glucose.....	10.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
DL-Methionine.....	0.02g
DL-Tryptophan.....	0.02g

L-Histidine·HCl.....	0.01g
Inositol.....	2.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyridoxine.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
p-Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.2mg
KI.....	0.1mg
CuSO ₄ ·5H ₂ O.....	0.04mg
Folic acid.....	2.0µg
Biotin.....	2.0µg
pH 4.5 ± 0.2 at 25°C	

Preparation of Yeast Nitrogen Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: To 100.0mL of yeast nitrogen base, add 10.0g of carbohydrate. Mix thoroughly. Filter sterilize. Aseptically distribute 0.5mL into tubes containing 4.5mL of sterile, distilled/deionized water.

Use: For the cultivation and differentiation of bacteria based on carbohydrate assimilation.

Wickerham Broth

Composition per 100.0mL:

KNO ₃	0.78g
Yeast carbon base.....	100.0mL
pH 4.5 ± 0.2 at 25°C	

Yeast Carbon Base:

Composition per liter:

Glucose.....	10.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
DL-Methionine.....	0.02g
DL-Tryptophan.....	0.02g
L-Histidine·HCl.....	0.01g
Inositol.....	2.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyridoxine.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
p-Aminobenzoic acid.....	0.2mg

Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
KI	0.1mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg

Preparation of Yeast Carbon Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: To 100.0mL of yeast carbon base, add 0.78g of KNO₃ (or peptone). Mix thoroughly. Filter sterilize. Aseptically distribute 0.5mL into tubes containing 4.5mL of sterile distilled/deionized water.

Use: For the cultivation and differentiation of bacteria based on nitrate assimilation.

Wickerham Broth with Raffinose

Composition per 100.0mL:

Raffinose	20.0g
Yeast nitrogen base	100.0mL

Yeast Nitrogen Base, 10X:

Composition per liter:

Glucose	10.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
DL-Methionine.....	0.02g
DL-Tryptophan.....	0.02g
L-Histidine-HCl	0.01g
Inositol	2.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine-HCl	0.4mg
Pyridoxine.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
KI	0.1mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg

pH 4.5 ± 0.2 at 25°C

Preparation of Yeast Nitrogen Base: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Preparation of Medium: To 100.0mL of yeast nitrogen base, add 20.0g of raffinose. Mix thoroughly. Filter sterilize. Aseptically distribute 0.5mL into tubes containing 4.5mL of sterile distilled/deionized water.

Use: For the cultivation and differentiation of bacteria based on carbohydrate assimilation.

Wilkins-Chalgren Anaerobe Agar

Composition per liter:

Agar	10.0g
Pancreatic digest of casein.....	10.0g
Gelatin peptone.....	10.0g
NaCl.....	5.0g
Yeast extract.....	5.0g
Glucose	1.0g
L-Arginine	1.0g
Sodium pyruvate.....	1.0g
Hemin	5.0mg
Menadione	0.5mg
Defibrinated blood	50.0mL
Tween 80.....	1.0mL

pH 7.1 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from Oxoid.

Preparation of Medium: Add components, except defibrinated blood, to distilled/deionized water and bring volume to 950.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C. Aseptically add 50.0mL of defibrinated blood. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of nonsporulating anaerobes.

Wilson Blair Base

Composition per liter:

Agar	30.0g
Proteose peptone No. 3	10.0g
Glucose	10.0g
Beef extract.....	5.0g
NaCl.....	5.0g
Selective reagent.....	70.0mL
Brilliant Green (1% solution)	4.0mL

pH 7.3 ± 0.2 at 25°C

Selective Reagent:

Composition per 100.0mL:

Solution 1.....	100.0mL
Solution 2.....	100.0mL
Solution 3.....	100.0mL
Solution 4.....	20.2mL

Preparation of Selective Reagent: Combine 100.0mL of solution 1, 100.0mL of solution 2,

100.0mL of solution 3, and 20.2mL of solution 4. Mix thoroughly. Gently heat to boiling until a slate-grey color develops. Cool to 50°C.

Solution 1:

Composition per 100.0mL:

NaHSO₃ 40.0g

Preparation of Solution 1: Add NaHSO₃ to 100.0mL of distilled/deionized water. Mix thoroughly.

Solution 2:

Composition per 100.0mL:

NaH₂PO₄ 21.0g

Preparation of Solution 2: Add NaH₂PO₄ to 100.0mL of distilled/deionized water. Mix thoroughly.

Solution 3:

Composition per 100.0mL:

Bismuth ammonium citrate.....12.5g

Preparation of Solution 3: Add bismuth ammonium citrate to 100.0mL of distilled/deionized water. Mix thoroughly.

Solution 4:

Composition per 20.2mL:

FeSO₄ 0.96g

Preparation of Solution 4: Add FeSO₄ to 20.0mL of distilled/deionized water. Add 0.2mL of concentrated HCl. Mix thoroughly.

Preparation of Medium: Add components, except selective reagent and Brilliant Green solution, to distilled/deionized water and bring volume to 976.0mL. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add selective reagent and Brilliant Green solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of *Salmonella*, especially *Salmonella typhi*.

Winge Melibiose Agar

Composition per liter:

Agar 15.0g

Melibiose 10.0g

Yeast extract.....3.0g

pH 7.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 7.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

WL Differential Agar (Wallerstein Laboratory Differential Agar)

Composition per liter:

Glucose 50.0g

Agar 20.0g

Pancreatic digest of casein.....5.0g

Yeast extract.....4.0g

KH₂PO₄.....0.55g

KCl.....0.425g

CaCl₂·2H₂O 0.125g

MgSO₄·7H₂O 0.125g

Bromcresol Green.....0.022g

Actidione (cycloheximide) 4.0mg

FeCl₃ 2.5mg

MnSO₄·4H₂O 2.5mg

pH 5.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differential cultivation of bacteria from industrial fermentation processes. Growth of yeasts and molds is inhibited.

WL Differential Medium (Wallerstein Laboratory Differential Medium)

Composition per liter:

Glucose 50.0g

Agar 20.0g

Pancreatic digest of casein.....5.0g

Yeast extract.....4.0g

KH₂PO₄.....0.55g

KCl.....0.425g

CaCl₂·2H₂O 0.125g

MgSO₄·7H₂O 0.125g

Bromcresol Green.....0.022g

Actidione (cycloheximide) 4.0mg

FeCl₃ 2.5mg

MnSO₄·4H₂O 2.5mg

pH 5.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differential cultivation of bacteria from industrial fermentation processes. Growth of yeasts and molds is inhibited.

WL Differential Medium (Wallerstein Laboratory Differential Medium)

Composition per liter:

Glucose	50.0g
Agar	20.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂ ·2H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g
Actidione (cycloheximide)	4.0mg
FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
Na ₂ CO ₃ (1% solution)	30.0mL
pH 6.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the differential cultivation of bacteria from industrial fermentation processes. Growth of yeasts and molds is inhibited.

WL Medium with Tomato Juice (Wallerstein Laboratory Medium with Tomato Juice)

Composition per liter:

Glucose	50.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g

FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
Tomato juice, canned, clarified	400.0mL
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of microorganisms from alcoholic mash.

WL Nutrient Agar (Wallerstein Laboratory Nutrient Agar)

Composition per liter:

Glucose	50.0g
Agar	20.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂ ·2H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g
FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the detection of bacteria and yeasts in industrial fermentation processes, particularly from beer processing. A medium for the determination of the microbiological flora in brewing and fermentation which can be made selective for bacteria with cycloheximide.

WL Nutrient Agar (Wallerstein Laboratory Nutrient Agar)

Composition per liter:

Glucose	50.0g
Agar	20.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂ ·2H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.125g

Bromcresol Green	0.022g
FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
Na ₂ CO ₃ (1% solution)	30.0mL
pH 6.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat with mixing and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the detection of bacteria and yeasts from industrial fermentation processes, particularly from beer processing.

WL Nutrient Broth (Wallerstein Laboratory Nutrient Broth)

Composition per liter:

Glucose	50.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g
FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
pH 5.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of yeasts, molds, and bacteria found in brewing and other industrial fermentation processes.

WL Nutrient Broth (Wallerstein Laboratory Nutrient Broth)

Composition per liter:

Glucose	50.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂	0.125g

MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g
FeCl ₃	2.5mg
MnSO ₄ ·4H ₂ O	2.5mg
Na ₂ CO ₃ (1% solution)	30.0mL
pH 6.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of yeasts, molds, and bacteria found in brewing and other industrial fermentation processes.

WL Nutrient Broth (Wallerstein Laboratory Nutrient Broth)

Composition per liter:

Glucose	50.0g
Pancreatic digest of casein	5.0g
Yeast extract	4.0g
FeCl ₃	2.5g
MnSO ₄ ·4H ₂ O	2.5g
KH ₂ PO ₄	0.55g
KCl	0.425g
CaCl ₂ ·2H ₂ O	0.125g
MgSO ₄ ·7H ₂ O	0.125g
Bromcresol Green	0.022g
pH 5.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the control of brewing and other fermentation processes.

Woods and Welton Agar

Composition per liter:

NaCl	23.4g
Casein hydrolysate	17.0g
Agar	15.0g
Glycerol	10.0g
Pancreatic digest of gelatin	5.0g
Glucose	5.0g
Papaic digest of soybean meal	3.0g
Beef extract	3.0g
Yeast extract	2.0g
Casamino acids, vitamin free	0.5g

Pancreatic digest of casein.....	0.5g
Na ₂ SO ₃	0.1g
pH 7.6 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.6. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Vibrio alginolyticus*.

Wort Agar

Composition per liter:

Agar	15.0g
Malt extract.....	15.0g
Maltose.....	12.75g
Dextrin	2.75g
Glycerol	2.35g
K ₂ HPO ₄	1.0g
NH ₄ Cl	1.0g
Pancreatic digest of gelatin.....	0.78g
pH 4.8 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Boil for 1 min with mixing. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Do not overheat, as this will result in hydrolysis of the agar. An additional 5.0g of agar can be used to make a firmer agar. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and enumeration of yeasts. The low pH of the agar selectively inhibits bacterial growth.

Wort Agar

Composition per liter:

Agar	25.0g
Yeast extract.....	1.0g
Wort solution.....	1.0L

Wort Solution:

Composition per liter:

Malt extract.....	110.0g

Preparation of Wort Solution: Add malt extract to distilled/deionized water and bring volume to 1000.0mL. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of yeasts and filamentous fungi.

Wort Broth

Composition per liter:

Yeast extract.....	1.0g
Wort solution.....	1.0L

Wort Solution:

Composition per liter:

Malt extract.....	110.0g

Preparation of Wort Solution: Add malt extract to distilled/deionized water and bring volume to 1000.0mL. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a wide variety of yeasts and filamentous fungi.

Wort Sucrose Agar

Composition per liter:

Agar	25.0g
Yeast extract.....	1.0g
Wort solution.....	500.0mL
Sucrose solution.....	500.0mL

Wort Solution:

Composition per 500.0mL:

Malt extract.....	55.0g

Preparation of Wort Solution: Add malt extract to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Sucrose Solution:

Composition per 500.0mL:

Sucrose.....	50.0g

Preparation of Sucrose Solution: Add sucrose to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Aspergillus oryzae*.

Wort Sucrose Broth

Composition per liter:

Yeast extract.....	1.0g

Wort solution.....	500.0mL
Sucrose solution.....	500.0mL

Wort Solution:**Composition per 500.0mL:**

Malt extract.....	55.0g

Preparation of Wort Solution: Add malt extract to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Sucrose Solution:**Composition per 500.0mL:**

Sucrose.....	50.0g

Preparation of Sucrose Solution: Add sucrose to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly.

Preparation of Medium: Combine components. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Aspergillus oryzae*.

XL Agar Base (Xylose Lysine Agar Base)

Composition per liter:

Agar	13.5g
Lactose	7.5g
Sucrose	7.5g
L-Lysine.....	5.0g
NaCl.....	5.0g
Xylose.....	3.5g
Yeast extract.....	3.0g
Phenol Red.....	0.08g
Thiosulfate-citrate solution.....	20.0mL
pH 7.5 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Thiosulfate-Citrate Solution:**Composition per 100.0mL:**

Na ₂ S ₂ O ₃	34.0g
Ferric ammonium citrate.....	4.0g

Preparation of Thiosulfate-Citrate Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except thiosulfate-citrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 14 psi pressure–118°C. Cool to 55°C. Aseptically add 20.0mL of the sterile thiosulfate-citrate solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and differentiation of enteric pathogens. Nonfermenting xylose/lactose/sucrose bacteria appear as red colonies. Xylose-fermenting, lysine-decarboxylating bacteria appear as red colonies. Xylose-fermenting, lysine-nondecaboxylating bacteria appear as opaque yellow colonies. Lactose or sucrose-fermenting bacteria appear as yellow colonies.

XL Agar Base

Composition per liter:

Agar	15.0g
Lactose	7.5g
Sucrose.....	7.5g
L-Lysine.....	5.0g
NaCl.....	5.0g
Xylose.....	3.75g
Yeast extract.....	3.0g
Phenol Red.....	0.08g
Thiosulfate-citrate solution.....	20.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Thiosulfate-Citrate Solution:**Composition per 100.0mL:**

Na ₂ S ₂ O ₃	34.0g
Ferric ammonium citrate.....	4.0g

Preparation of Thiosulfate-Citrate Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly.

Preparation of Medium: Add components, except thiosulfate-citrate solution, to distilled/deionized water and bring volume to 980.0mL. Mix thoroughly. Gently heat while stirring and bring to boiling. Distribute into tubes or flasks. Autoclave for 10 min at 14 psi pressure–118°C. Cool to 55°C. Aseptically add 20.0mL of the sterile thiosulfate-citrate solution. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and differentiation of enteric pathogens. Nonfermenting xylose/lactose/sucrose bacteria appear as red colonies. Xylose-fermenting, lysine-decarboxylating bacteria appear as red colonies. Xylose-fermenting, lysine-nondecaboxylating bacteria appear as opaque yellow colonies. Lactose or sucrose-fermenting bacteria appear as yellow colonies.

XLD Agar (Xylose Lysine Deoxycholate Agar) (BAM M179)

Composition per liter:

Agar	15.0g

Lactose	7.5g
Sucrose	7.5g
Na ₂ S ₂ O ₃	6.8g
L-Lysine	5.0g
NaCl	5.0g
Xylose	3.75g
Yeast extract	3.0g
Sodium deoxycholate	2.5g
Ferric ammonium citrate	0.8g
Phenol Red	0.08g

pH 7.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Do not overheat. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes. Plates should be poured as soon as possible to avoid precipitation.

Use: For the isolation and differentiation of enteric pathogens, especially *Shigella* and *Providencia* species. Nonfermenting xylose/lactose/sucrose bacteria appear as red colonies. Xylose-fermenting, lysine-decarboxylating bacteria appear as red colonies. Xylose-fermenting, lysine-nondecarboxylating bacteria appear as opaque yellow colonies. Lactose or sucrose-fermenting bacteria appear as yellow colonies.

Xylose Sodium Deoxycholate Citrate Agar

Composition per liter:

Agar	12.0g
Xylose	10.0g
Sodium citrate	5.0g
Na ₂ S ₂ O ₃ ·5H ₂ O	5.0g
Beef extract	5.0g
Peptone	5.0g
NaCl	2.5g
Sodium deoxycholate	2.5g
Ferric ammonium citrate	1.0g
Neutral Red (1% solution)	2.5mL

pH 7.5 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling for 20 sec. Do not autoclave. Cool to 45°–50°C. Pour into sterile Petri dishes.

Use: For the cultivation of *Salmonella* species and some *Shigella* species.

Xylose YP Agar

(Xylose Yeast Extract Peptone Agar)

Composition per liter:

CaCO ₃	20.0g
Agar	15.0g
Xylose	10.0g
Yeast extract	10.0g
Peptone	10.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.01g
FeSO ₄ ·7H ₂ O	0.01g
NaCl	0.01g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into screw-capped test tubes. Autoclave for 15 min at 15 psi pressure–121°C. Adjust pH to 6.8. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Lactobacillus vaccिनosterсus* and other microorganisms that utilize xylose as a carbon source.

Xylose YP Broth

(Xylose Yeast Extract Peptone Broth)

Composition per liter:

Xylose	10.0g
Yeast extract	10.0g
Peptone	10.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.01g
FeSO ₄ ·7H ₂ O	0.01g
NaCl	0.01g

pH 6.8 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into screw-capped test tubes. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Lactobacillus vaccिनosterсus* and other microorganisms that utilize xylose as a carbon source.

Y1 Adrenal Cell Growth Medium

Composition per 101.0mL:

Ham's F-10 medium	90.0mL
Fetal bovine serum	10.0mL
Penicillin-streptomycin solution	1.0mL

pH 7.0 ± 0.2 at 25°C

Ham's F-10 Medium:**Composition per liter:**

NaCl.....	7.4g
NaHCO ₃	1.2g
Glucose	1.1g
NaH ₂ PO ₄ ·H ₂ O.....	0.29g
KCl.....	0.28g
L-Arginine·HCl.....	0.21g
L-Glutamine.....	0.15g
MgSO ₄ ·7H ₂ O	0.15g
Sodium pyruvate.....	0.11g
KH ₂ PO ₄	0.08g
CaCl ₂ ·2H ₂ O.....	0.04g
L-Cystine·2HCl.....	0.04g
L-Histidine·HCl·H ₂ O	0.02g
L-Lysine·HCl	0.02g
L-Asparagine·H ₂ O.....	0.01g
L-Aspartic Acid	0.01g
L-Glutamic acid.....	0.01g
L-Leucine.....	0.01g
L-Proline.....	0.01g
L-Serine	0.01g
L-Alanine.....	8.9mg
Glycine.....	7.5mg
D-Phenylalanine	5.0mg
L-Methionine	4.5mg
Hypoxanthine.....	4.1mg
L-Threonine.....	3.6mg
L-Valine	3.5mg
L-Isoleucine.....	2.6mg
L-Tyrosine.....	1.8mg
Vitamin B ₁₂	1.4mg
Folic acid.....	1.3mg
Phenol Red.....	1.2mg
Thiamine·HCl	1.0mg
FeSO ₄ ·7H ₂ O.....	0.8mg
Choline chloride.....	0.7mg
D-Calcium pantothenate.....	0.7mg
Thymidine.....	0.7mg
Niacinamide.....	0.6mg
L-Tryptophan.....	0.6mg
Isoinositol.....	0.5mg
Riboflavin.....	0.4mg
Lipoic acid.....	0.2mg
Pyridoxine·HCl	0.2mg
ZnSO ₄ ·7H ₂ O	0.03mg
Biotin	0.02mg
CuSO ₄ ·5H ₂ O	3.0μg

Preparation of Ham's F-10 Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Penicillin-Streptomycin Solution:**Composition per 100.0mL:**

Streptomycin.....	0.5g
Penicillin G	500,000U

Preparation of Penicillin-Streptomycin Solution: Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine components. Filter sterilize. Store at 4-5°C.

Use: For the cultivation of Y-1 mouse adrenal tissue culture cells used for the detection of heat-labile toxin (LT) produced by enterotoxigenic strains of *Escherichia coli*. LT causes the conversion of elongated fibroblast-like cells into round, refractile cells.

Y1 Adrenal Cell Growth Medium**Composition per 580.0mL:**

Ham's F-10 medium	500.0mL
Fetal bovine serum.....	75.0mL
Penicillin-streptomycin solution.....	5.0mL
pH 7.0 ± 0.2 at 25°C	

Ham's F-10 Medium:**Composition per liter:**

NaCl.....	7.4g
NaHCO ₃	1.2g
Glucose	1.1g
NaH ₂ PO ₄ ·H ₂ O.....	0.29g
KCl.....	0.28g
L-Arginine·HCl.....	0.21g
L-Glutamine	0.15g
MgSO ₄ ·7H ₂ O	0.15g
Sodium pyruvate.....	0.11g
KH ₂ PO ₄	0.08g
CaCl ₂ ·2H ₂ O	0.04g
L-Cystine·2HCl	0.04g
L-Histidine·HCl·H ₂ O	0.02g
L-Lysine·HCl.....	0.02g
L-Asparagine·H ₂ O.....	0.01g
L-Aspartic Acid	0.01g
L-Glutamic acid.....	0.01g
L-Leucine	0.01g
L-Proline.....	0.01g
L-Serine	0.01g
L-Alanine.....	8.9mg
Glycine.....	7.5mg
D-Phenylalanine	5.0mg
L-Methionine	4.5mg
Hypoxanthine.....	4.1mg
L-Threonine.....	3.6mg
L-Valine	3.5mg
L-Isoleucine.....	2.6mg
L-Tyrosine	1.8mg

Vitamin B ₁₂	1.4mg
Folic acid.....	1.3mg
Phenol Red.....	1.2mg
Thiamine-HCl.....	1.0mg
FeSO ₄ ·7H ₂ O.....	0.8mg
Choline chloride.....	0.7mg
D-Calcium pantothenate.....	0.7mg
Thymidine.....	0.7mg
Niacinamide.....	0.6mg
L-Tryptophan.....	0.6mg
Inositol.....	0.5mg
Riboflavin.....	0.4mg
Lipoic acid.....	0.2mg
Pyridoxine-HCl.....	0.2mg
ZnSO ₄ ·7H ₂ O.....	0.03mg
Biotin.....	0.02mg
CuSO ₄ ·5H ₂ O.....	3.0μg

Preparation of Ham's F-10 Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly.

Penicillin-Streptomycin Solution:

Composition per 100.0mL:

Streptomycin.....	0.5g
Penicillin G.....	500,000U

Preparation of Penicillin-Streptomycin Solution:

Add components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Aseptically combine components. Filter sterilize. Store at 4°–5°C.

Use: For the cultivation of Y-1 mouse adrenal tissue culture cells used for the detection of cholera enterotoxin (CT) produced by enterotoxigenic strains of *Vibrio cholerae* or *Vibrio mimicus*. CT causes the conversion of elongated fibroblast-like cells into round, refractile cells. Also for the detection of toxin producing strains of *Escherichia coli*.

YC Agar without Tryptophan

Composition per liter:

Solution A.....	500.0mL
Solution B.....	500.0mL

Solution A:

Composition per 500.0mL:

Glucose.....	20.0g
Succinic acid.....	10.0g
NaOH.....	6.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	0.6g
MgSO ₄ ·7H ₂ O.....	0.3g
Adenine.....	0.1g
L-Arginine.....	0.1g

L-Cysteine.....	0.1g
L-Leucine.....	0.1g
L-Lysine.....	0.1g
L-Threonine.....	0.1g
Uracil.....	0.1g
L-Aspartic acid.....	0.05g
L-Histidine.....	0.05g
L-Isoleucine.....	0.05g
L-Methionine.....	0.05g
L-Phenylalanine.....	0.05g
L-Proline.....	0.05g
L-Serine.....	0.05g
L-Tyrosine.....	0.05g
L-Valine.....	0.05g
NaCl.....	0.05g
CaCl ₂ ·2H ₂ O.....	0.05g
DL-Methionine.....	0.01g
L-Histidine-HCl.....	0.005g
Inositol.....	1.0mg
KI.....	0.5mg
H ₃ BO ₃	0.3mg
ZnSO ₄ ·7H ₂ O.....	0.2mg
MnSO ₄ ·4H ₂ O.....	0.2mg
Thiamine-HCl.....	0.2mg
Pyridoxine-HCl.....	0.2mg
Niacin.....	0.2mg
Calcium pantothenate.....	0.2mg
p-Aminobenzoic acid.....	0.1mg
Riboflavin.....	0.1mg
FeCl ₃	0.1mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.1mg
CuSO ₄ ·5H ₂ O.....	0.02mg
Folic acid.....	1.0μg
Biotin.....	1.0μg

pH 5.7 ± 0.2 at 25°C

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Adjust pH to 5.6–5.7. Filter sterilize. Warm to 50°–55°C.

Solution B:

Composition per 500.0mL:

Agar, noble.....	20.0g

Preparation of Solution B: Add agar to distilled/deionized water and bring volume to 500.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°–55°C.

Preparation of Medium: Aseptically mix 500.0mL of solution A and 500.0mL of solution B. Mix thoroughly. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

YC Medium without Tryptophan**Solution A:****Composition per 500.0mL:**

Glucose	20.0g
Succinic acid	10.0g
NaOH	6.0g
(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
Adenine	0.1g
L-Arginine	0.1g
L-Cysteine	0.1g
L-Leucine	0.1g
L-Lysine	0.1g
L-Threonine	0.1g
Uracil	0.1g
L-Aspartic acid	0.05g
L-Histidine	0.05g
L-Isoleucine	0.05g
L-Methionine	0.05g
L-Phenylalanine	0.05g
L-Proline	0.05g
L-Serine	0.05g
L-Tyrosine	0.05g
L-Valine	0.05g
Inositol	2.0mg
KI	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine·HCl	0.4mg
Pyroxidine·HCl	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid	2.0μg
Biotin	2.0μg

Preparation of Solution A: Add components to distilled/deionized water and bring volume to 500.0mL. Adjust pH to 5.6–5.7. Filter sterilize. Warm to 50°–55°C.

Solution B:**Composition per 500.0mL:**

Agar, noble	20.0g

Preparation of Solution B: Add agar to distilled/deionized water and bring volume to 500.0mL. Gen-

tly heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50–55°C.

Preparation of Medium: Aseptically combine 500.0mL of solution A and 500.0mL of solution B. Pour into sterile Petri dishes or distribute into sterile tubes.

Use: For the cultivation of *Saccharomyces cerevisiae*.

Yeast Ascospore Agar**Composition per liter:**

Agar	30.0g
Potassium acetate	10.0g
Yeast extract	2.5g
Glucose	1.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation and observation of ascospore formation of yeast.

**Yeast Carbon Base, 10X
(Wickerham Carbon Base Broth)****Composition per liter:**

Glucose	10.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl	0.1g
CaCl ₂ ·2H ₂ O	0.1g
DL-Methionine	0.02g
DL-Tryptophan	0.02g
L-Histidine·HCl	0.01g
Inositol	2.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine·HCl	0.4mg
Pyridoxine	0.4mg
Niacin	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
KI	0.1mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic Acid	2.0μg
Biotin	2.0μg

pH 5.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Use: Used as a base to which different nitrogen sources may be added. For the cultivation and differentiation of bacteria based on their ability to utilize diverse added nitrogen sources.

Yeast Dextrose Agar

Composition per liter:

Agar	15.0g
Glucose	10.0g
Yeast extract	10.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Adjust pH to 7.0. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of a variety of heterotrophic microorganisms.

Yeast Extract Agar (BAM M181)

Composition per liter:

Agar	15.0g
Proteose peptone	10.0g
NaCl	5.0g
Yeast extract	3.0g

pH 7.3 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of heterotrophic microorganisms.

Yeast Extract Malt Extract Glucose Agar

Composition per liter:

Agar	20.0g
Glucose	10.0g
Neopeptone	5.0g
Malt extract	3.0g
Yeast extract	3.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min

at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and cultivation of yeasts.

Yeast Extract Peptone Starch Agar

Composition per liter:

Agar	18.0g
Soluble starch	10.0g
Peptone	10.0g
CaCO ₃	5.0g
Sodium acetate	5.0g
Yeast extract	3.0g
KH ₂ PO ₄	0.5g
K ₂ HPO ₄	0.5g
MgSO ₄ ·7H ₂ O	0.3g
Sodium citrate	0.027g
NaCl	0.01g
MnSO ₄ ·5H ₂ O	0.01g
CuSO ₄ ·5H ₂ O	1.0mg
CoCl ₂ ·6H ₂ O	1.0mg
FeSO ₄ ·7H ₂ O	1.0mg

Preparation of Medium: Add components to tap water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus* species that utilize starch as a carbon source.

Yeast Extract Sodium Lactate Medium

Composition per liter:

Agar	15.0g
Pancreatic digest of casein	10.0g
Yeast extract	10.0g
Sodium lactate	10.0g
KH ₂ PO ₄	2.5g
MnSO ₄	5.0mg

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation, cultivation, and maintenance of *Propionibacterium* species.

Yeast Extract Sucrose Agar (YESA) (ATCC Medium 2125)

Composition per liter:

Sucrose	20.0g

Agar	15.0g
Yeast extract.....	4.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
pH 6.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of various fungi.

Yeast Extract Sucrose Agar

Composition per liter:

Sucrose.....	150.0g
Agar	20.0g
Yeast extract.....	20.0g
MgSO ₄ ·7H ₂ O	0.5g
Trace metal solution.....	1.0mL
pH 6.5 ± 0.2 at 25°C	

Trace Metal Solution:

Composition per 100.0mL:

ZnSO ₄ ·7H ₂ O	1.0g
CuSO ₄ ·5H ₂ O	0.5g

Preparation of Trace Metal Solution: Add components to 100.0mL distilled/deionized water. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.5. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of various fungi.

Yeast Fermentation Broth

Composition per liter:

Carbohydrate.....	10.0g
Pancreatic digest of gelatin.....	7.5g
Yeast extract.....	5.5g
Bromocresol Purple	16.0mg

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into test tubes, each containing an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For fermentation tests of specific carbohydrates used in the characterization and identification of yeasts. Gas accumulation in the Durham tube and a color change of the medium to yellow indicates carbohydrate fermentation.

Yeast Fermentation Medium

Composition per liter:

Peptone	7.5g
Yeast extract.....	4.5g
Bromthymol Blue (1.6% solution)	1.0mL
Carbohydrate solution.....	1.0mL

Carbohydrate Solution:

Composition per 10.0mL:

Carbohydrate.....	0.6g

Preparation of Carbohydrate Solution: Add carbohydrate to distilled/deionized water and bring volume to 10.0mL. Glucose, maltose, lactose, galactose, or trehalose may be used. If raffinose is used, prepare a 12% solution. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except carbohydrate solution, to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute in 2.0mL volumes into test tubes that contain an inverted Durham tube. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. Aseptically add 1.0mL of sterile carbohydrate solution. Mix thoroughly.

Use: For the cultivation and differentiation of yeast based on carbohydrate fermentation patterns. Yeasts that can ferment a specific carbohydrate turn the medium yellow.

Yeast Glucose Agar

Composition per liter:

Agar	15.0g
Pancreatic digest of gelatin.....	7.75g
Beef extract.....	4.75g
Yeast extract.....	2.5g
K ₂ HPO ₄	2.5g
Glucose	1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of a wide variety of bacteria.

Yeast Glucose Broth

Composition per liter:

Pancreatic digest of gelatin.....	7.75g
Beef extract.....	4.75g
Yeast extract.....	2.5g
K ₂ HPO ₄	2.5g
Glucose.....	1.0g

pH 7.0 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Staphylococcus caseolyticus*.

Yeast Malt Extract Broth with Glucose (YM Broth with Glucose)

Composition per liter:

Glucose.....	290.0g
Peptone.....	5.0g
CaCO ₃	5.0g
Yeast extract.....	3.0g
Malt extract.....	3.0g

pH 6.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. The medium may be rendered selective by adjusting the pH to 3.0–4.0 at 45°–55°C or by the addition of antibiotics.

Use: For the cultivation of yeasts and filamentous fungi.

Yeast Nitrogen Base

Composition per liter:

(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
DL-Methionine.....	0.02g
DL-Tryptophan.....	0.02g
L-Histidine·HCl.....	0.01g
Inositol.....	2.0mg
KI.....	1.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine·HCl.....	0.4mg
Pyroxidine·HCl.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg

p-Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.2mg
CuSO ₄ ·5H ₂ O.....	0.04mg
Folic acid.....	2.0μg
Biotin.....	2.0μg

pH 5.5 ± 0.2 at 25°C

Source: This medium is available as a premixed powder from BD Diagnostic Systems.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Alternately for carbon assimilation tests, prepare a 10× concentrated solution by adding components to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Prepare a carbohydrate solution by adding 0.5g of carbohydrate to 90.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize. Aseptically add 0.5mL of the 10× concentrated solution to 4.5mL of the filter-sterilized carbohydrate solution. Mix thoroughly.

Use: For carbohydrate assimilation tests in the characterization and identification of yeasts.

Yeast Synthetic Medium with 5'-dTMP

Composition per liter:

Glucose.....	20.0g
Agar.....	15.0g
Yeast nitrogen base without amino acids.....	6.7g
Casamino acids without vitamins.....	2.0g
2'-Deoxythymidine-5'-monophosphate.....	7.2mg

Preparation of Medium: Add glucose, yeast nitrogen base without amino acids, casamino acids without vitamins, and 2'-deoxythymidine-5'-monophosphate to distilled/deionized water and bring volume to 200.0mL. Mix thoroughly. Filter sterilize. Add agar to distilled/deionized water and bring volume to 800.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically combine the two sterile solutions. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

Yeast Synthetic Minimal Medium

Composition per liter:

D-Glucose.....	20.0g
Agar.....	15.0g

(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
Inositol	2.0mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O	0.4mg
MnSO ₄ ·4H ₂ O	0.4mg
Thiamine-HCl	0.4mg
Pyridoxine-HCl	0.4mg
Niacin.....	0.4mg
Calcium pantothenate	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O	0.2mg
KI	0.1mg
CuSO ₄ ·5H ₂ O	0.04mg
Folic acid.....	2.0μg
Biotin	2.0μg
pH 5.6 ± 0.2 at 25°C	

Preparation of Medium: Add agar to 900.0mL of distilled/deionized water. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 45°–50°C. In a separate flask, add remaining components to 100.0mL of distilled/deionized water. Mix thoroughly. Filter sterilize. Aseptically combine the two sterile solutions. Mix thoroughly. Pour into sterile Petri dishes.

Use: For the cultivation of a wide variety of heterotrophic microorganisms.

Yeast Synthetic Minimal Medium

Composition per liter:

Glucose	20.0g
Agar	15.0g
Yeast nitrogen base without amino acids.....	6.7g

Preparation of Medium: Add glucose and yeast nitrogen base without amino acids to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize. Add agar to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically combine the two sterile solutions. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and maintenance of *Pichia angusta* and *Saccharomyces cerevisiae*.

Yeast Tryptone Medium

Composition per liter:

Pancreatic digest of casein.....	10.0g
NaCl.....	10.0g
Yeast extract.....	5.0g
pH 7.0 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of *Escherichia coli*.

Yeast Tryptone Starch Medium

Composition per liter:

Agar	15.0g
Soluble starch.....	10.0g
Yeast extract.....	5.0g
Pancreatic digest of casein.....	5.0g
KH ₂ PO ₄	2.0g
CaCl ₂ ·2H ₂ O	0.5g
MnCl ₂ ·4H ₂ O	0.5g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Bacillus circulans*.

Yeastrel Agar

Composition per liter:

Agar	15.0g
Peptone	9.5g
Yeastrel	7.0g
Lab-lemco (meat extract).....	5.0g
NaCl.....	5.0g
pH 7.0 ± 0.2 at 25°C	

Source: Lab-lemco is available from Oxoid. Yeastrel is produced by Mapleton's Foods Ltd., Moss Street, Liverpool and is available from health food shops.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of *Aeromonas salmonicida*.

YEP Galactose Agar**Composition per liter:**

Agar	20.0g
Galactose.....	20.0g
Peptone.....	20.0g
Yeast extract.....	10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of heterotrophic microorganisms.

YEP Galactose Medium**Composition per liter:**

Agar	20.0g
Galactose.....	20.0g
Peptone.....	20.0g
Yeast extract.....	10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae* and *Yarrowia lipolytica*.

YEPD-FA Medium**Composition per liter:**

Agar	20.0g
Glucose	20.0g
Peptone.....	20.0g
Yeast extract.....	10.0g
Tween 40.....	10.0g
KH ₂ PO ₄	5.0g
K ₂ HPO ₄	5.0g
Myristic acid	70.0mg
Palmitic acid	70.0mg
Stearic acid.....	70.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

YEPD Inositol Agar**Composition per liter:**

Glucose	20.0g
Mycological peptone	20.0g
Agar	15.0g
Yeast extract.....	10.0g
Inositol	180.0mg

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

YEPD Medium**Composition per liter:**

Agar	20.0g
Glucose	20.0g
Peptone	20.0g
Yeast extract.....	10.0g

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of heterotrophic microorganisms.

YEPD Medium with Heme**Composition per liter:**

Agar	20.0g
Glucose	20.0g
Peptone	20.0g
Yeast extract.....	10.0g
Hemin stock solution	20.0mL

Hemin Stock Solution:**Composition per 10mL:**

NaOH.....	0.04g
Hemin chloride	65.0mg
Ethanol (50% solution).....	100.0mL

Preparation of Hemin Stock Solution: Combine components and mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

***Yersinia* Selective Agar Base**

Composition per liter:

Mannitol.....	20.0g
Peptone.....	17.0g
Agar	12.5g
Proteose peptone.....	3.0g
Yeast extract.....	2.0g
Sodium pyruvate.....	2.0g
NaCl.....	1.0g
Sodium deoxycholate.....	0.5g
MgSO ₄ ·7H ₂ O	0.01g
Neutral Red.....	0.03g
Crystal Violet.....	1.0mg
Selective supplement	6.0mL
pH 7.4 ± 0.2 at 25°C	

Source: This medium is available as a premixed powder from BD Diagnostic Systems and Oxoid.

Selective Supplement:

Composition per 6.0mL:

Cefsulodin.....	15.0mg
Irgasan.....	4.0mg
Novobiocin.....	2.5mg
Ethanol	2.0mL

Preparation of Selective Supplement: Aseptically add components to 4.0mL of distilled/deionized water and 2.0mL of ethanol. Mix thoroughly.

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add selective supplement. Mix thoroughly. Pour into sterile Petri dishes or leave in tubes.

Use: For the isolation and enumeration of *Yersinia enterocolitica* from foods.

YGC Medium

**(Yeast Extract Glucose Citrate Medium)
(ATCC Medium 216)**

Composition per liter:

Beef extract.....	10.0g
Glucose	10.0g
Peptone.....	10.0g
Ammonium citrate.....	5.0g
Yeast extract.....	5.0g
Sodium acetate.....	2.0g
Tween 80.....	1.0g

MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
pH 6.5 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the isolation and cultivation of *Leuconostoc* species.

YGC Medium with Cysteine

(Yeast Extract Glucose

Citrate Medium with Cysteine)

Composition per liter:

Glucose	10.0g
Peptone	10.0g
Beef extract.....	10.0g
Yeast extract.....	5.0g
Ammonium citrate.....	5.0g
Sodium acetate.....	2.0g
Tween 80.....	1.0g
MgSO ₄ ·7H ₂ O	0.2g
MnSO ₄ ·4H ₂ O	0.05g
L-Cysteine·HCl·H ₂ O solution	10.0mL
pH 6.5 ± 0.2 at 25°C	

L-Cysteine·HCl Solution:

Composition per 10.0mL:

L-Cysteine·HCl·H ₂ O	0.5g

Preparation of L-Cysteine·HCl Solution: Add L-cysteine·HCl·H₂O to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except L-cysteine·HCl·H₂O, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add L-cysteine hydrochloride solution.

Use: For the cultivation and maintenance of *Leuconostoc mesenteroides*.

YM-1L Broth

Composition per liter:

Sodium lactate	30.0g
Mycological peptone	10.0g
Succinic acid.....	10.0g
NaOH.....	6.0g
Yeast extract.....	5.0g
Adenine.....	0.01g
Uracil	0.01g
Yeast nitrogen base solution	100.0mL

Yeast Nitrogen Base Solution:**Composition per 100mL:**

(NH ₄) ₂ SO ₄	5.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O.....	0.5g
NaCl.....	0.1g
CaCl ₂ ·2H ₂ O.....	0.1g
DL-Methionine.....	0.02g
DL-Tryptophan.....	0.02g
L-Histidine-HCl.....	0.01g
Inositol.....	2.0mg
KI.....	0.1mg
H ₃ BO ₃	0.5mg
ZnSO ₄ ·7H ₂ O.....	0.4mg
MnSO ₄ ·4H ₂ O.....	0.4mg
Thiamine-HCl.....	0.4mg
Pyroxidine-HCl.....	0.4mg
Niacin.....	0.4mg
Calcium pantothenate.....	0.4mg
<i>p</i> -Aminobenzoic acid.....	0.2mg
Riboflavin.....	0.2mg
FeCl ₃	0.2mg
Na ₂ MoO ₄ ·4H ₂ O.....	0.2mg
CuSO ₄ ·5H ₂ O.....	0.04mg
Folic acid.....	2.0μg
Biotin.....	2.0μg

Preparation of Yeast Nitrogen Base Solution:

Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except yeast nitrogen base solution, to distilled/deionized water and bring volume to 900.0mL. Mix thoroughly. Autoclave for 15 min at 15 psi pressure–121°C. Aseptically add 1.0L of sterile yeast nitrogen base solution. Mix thoroughly. Aseptically distribute into sterile tubes or flasks.

Use: For the cultivation of *Saccharomyces cerevisiae*.

YMF Agar**Composition per liter:**

Agar.....	20.0g
Peptone.....	5.0g
Sugar, brown.....	3.0g
Malt extract.....	3.0g
Yeast extract.....	3.0g

pH 6.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of various fungi.

YMF Broth**Composition per liter:**

Peptone.....	5.0g
Sugar, brown.....	3.0g
Malt extract.....	3.0g
Yeast extract.....	3.0g

pH 6.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Adjust pH to 6.2. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C.

Use: For the cultivation of various fungi.

YPDP Medium with 5'-TMP**Composition per liter:**

Glucose.....	20.0g
Peptone.....	20.0g
Agar.....	15.0g
Yeast extract.....	10.0g
KH ₂ PO ₄	1.5g
Thymidine-5'-monophosphate solution.....	10.0mL

Thymidine-5'-Monophosphate Solution:**Composition per 10mL:**

Thymidine-5'-monophosphate.....	100.0mg

Preparation of Thymidine-5'-Monophosphate Solution: Add thymidine-5'-monophosphate to distilled/deionized water and bring volume to 10.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except thymidine-5'-monophosphate solution, to distilled/deionized water and bring volume to 990.0mL. Mix thoroughly. Gently heat and bring to boiling. Autoclave for 15 min at 15 psi pressure–121°C. Cool to 50°C. Aseptically add 10.0mL of sterile thymidine-5'-monophosphate solution. Mix thoroughly. Pour into sterile Petri dishes or aseptically distribute into sterile tubes.

Use: For the cultivation and maintenance of *Saccharomyces cerevisiae*.

YPG Agar with 2% NaCl**Composition per liter:**

Agar.....	20.0g
Glucose.....	20.0g
NaCl.....	20.0g
Peptone.....	10.0g
Yeast extract.....	10.0g

pH 5.2 ± 0.2 at 25°C

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L.

Mix thoroughly. Adjust pH to 5.2. Gently heat and bring to boiling. Distribute into tubes or flasks. Autoclave for 15 min at 15 psi pressure–121°C. Pour into sterile Petri dishes or leave in tubes.

Use: For the cultivation of a variety of yeasts and other fungi

Z-Agar

Composition per liter:

Agar	16.0
K ₂ HPO ₄	5.0g
K ₂ SO ₄	2.0g
KH ₂ PO ₄	1.0g
MgSO ₄ ·7H ₂ O	0.05g
Acetamide solution	40.0mL
pH 7.2 ± 0.2 at 25°C	

Acetamide Solution:

Composition per 100.0mL:

Acetamide	10.0g

Preparation of Acetamide Solution: Add acetamide to distilled/deionized water and bring volume to 100.0mL. Mix thoroughly. Filter sterilize.

Preparation of Medium: Add components, except acetamide solution, to distilled/deionized water

and bring volume to 960.0mL. Mix thoroughly. Autoclave for 20 min at 15 psi pressure–121°C. Cool to 70°C. Aseptically add 40.0mL sterile acetamide solution. Mix thoroughly. Pour into sterile Petri dishes. Dry plates at 37°C for 30 min.

Use: For the isolation of *Pseudomonas aeruginosa* from milk.

Z-Broth

Composition per liter:

Acetamide	5.0g
K ₂ HPO ₄	5.0g
KH ₂ PO ₄	3.0g
KNO ₃	1.0g
K ₂ S ₄ O ₆	1.0g
MgSO ₄ ·7H ₂ O	0.05g
pH 7.2 ± 0.2 at 25°C	

Preparation of Medium: Add components to distilled/deionized water and bring volume to 1.0L. Mix thoroughly. Gently heat and bring to boiling. Cool. Aseptically distribute 10.0mL volumes into test tubes containing inverted Durham tubes. Heat for 15 min at 0 psi pressure–100°C.

Use: For the cultivation of *Pseudomonas aeruginosa* from milk.

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Composition of Media

Media for the cultivation of microorganisms contain the substances necessary to support the growth of microorganisms. Due to the diversity of microorganisms and their diverse metabolic pathways, there are numerous media. Even slight differences in the composition of a medium can result in dramatically different growth characteristics of microorganisms.

The composition section of each medium describes the ingredients that make up the medium, their amounts, and the pH. It lists those ingredients in order of decreasing amount. Solids are listed first showing the weights to be added, followed by liquids showing the volumes to be included in the medium. The composition uses generic terms where these are applicable. For example, pancreatic digest of casein is marketed by various manufacturers as trypticase, tryptone, and other commercial product names. While there may well be differences between these products, such differences are undefined. Variations also occur between batches of products produced as digests of animal tissues.

Agars

Agar is the most common solidifying agent used in microbiological media. Agar is a polysaccharide extract from marine algae. It melts at 84°C and solidifies at 38°C. Agar concentrations of 15.0g/L typically are used to form solid media. Lower concentrations of 7.5 to 10.0g/L are used to produce soft agars or semisolid media. Below are some agars used as solidifying agents in various media.

Agar Bacteriological (Agar No. 1)

An agar with low calcium and magnesium. Available from Oxoid.

Agar, Bacto

A purified agar with reduced pigmented compounds, salts, and extraneous matter. Available from BD Diagnostic Systems.

Agar, BiTek™

Agar prepared as a special technical grade. Available from BD Diagnostic Systems.

Agar, Flake

A technical grade agar. Available from BD Diagnostic Systems.

Agar, Grade A

A select grade agar containing minerals. Available from BD Diagnostic Systems.

Agar, Granulated

A high grade granulated agar that has been filtered, decolorized, and purified. Available from BD Diagnostic Systems.

Agarose

A low sulfate neutral gelling fraction of agar that is a complex galactose polysaccharide of near neutral charge.

Agar, Purified

A very high grade agar that has been filtered, decolorized, and purified by washing and extraction of refined agars. It has reduced mineral content. Available from BD Diagnostic Systems.

Agar Technical (Agar No. 3)

A technical grade agar. Available from BD Diagnostic Systems and Oxoid.

Ionagar

A purified agar. Available from Oxoid.

Noble Agar

An agar that has been extensively washed and is essentially free of impurities. Available from BD Diagnostic Systems.

Purified Agar

An agar that has been extensively washed and extracted with water and organic solvent. Available from BD Diagnostic Systems and Oxoid.

Peptones

Many complex media, that is, media in which not all the specific chemical components are known, contain peptones as the source of nitrogen. Peptones are hydrolyzed proteins formed by enzymatic or acidic digestion. Casein most often is used as the protein substrate for forming peptones, but other substances, such as soybean meal, also are commonly employed.

Below is a list of some of the peptones that are used as ingredients in various media.

Acidase™ Peptone

A hydrochloric acid hydrolysate of casein. It has a nitrogen content of 8% and is deficient in cystine and tryptophan. Available from BD Diagnostic Systems.

Bacto Casitone

A pancreatic digest of casein. Available from BD Diagnostic Systems.

Bacto Peptamin

A peptic digest of animal tissues. Available from BD Diagnostic Systems.

Bacto Peptone

An enzymatic digest of animal tissues. It has a high concentration of low molecular weight peptones and amino acids. Available from BD Diagnostic Systems.

Bacto Proteose Peptone

An enzymatic digest of animal tissues. It has a high concentration of high molecular weight peptones. Available from BD Diagnostic Systems.

Bacto Soytone

A enzymatic hydrolysate of soybean meal. Available from BD Diagnostic Systems.

Bacto Tryptone

A pancreatic digest of casein. Available from BD Diagnostic Systems.

Bacto Tryptose

An enzymatic hydrolysate containing numerous peptides including those of higher molecular weights. Available from BD Diagnostic Systems.

Biosate™ Peptone

A hydrolysate of plant and animal proteins. Available from BD Diagnostic Systems.

Casein Hydrolysate

A hydrolysate of casein prepared with hydrochloric acid digestion under pressure and neutralized with sodium hydroxide. It contains total nitrogen of 7.6% and NaCl of 28.3%. Available from Oxoid.

Gelatone

A pancreatic digest of gelatin. Available from BD Diagnostic Systems.

Gelysate™ Peptone

A pancreatic digest of gelatin deficient in cystine and tryptophan and which has a low carbohydrate content. Available from Oxoid.

Lactoalbumin Hydrolysate

A pancreatic digest of lactoalbumin, a milk whey protein. It has high levels of amino acids. It contains total nitrogen of 11.9% and NaCl of 1.4%. Available from BD Diagnostic Systems and Oxoid.

Liver Digest Neutralized

A papaic digest of liver that contains total nitrogen of 11.0% and NaCl of 1.6%. Available from Oxoid.

Mycological Peptone

A peptone that contains total nitrogen of 9.5% and NaCl of 1.1%. Available from Oxoid.

Myosate™ Peptone

A pancreatic digest of heart muscle. Available from BD Diagnostic Systems.

Neopeptone

An enzymatic digest of protein. Available from BD Diagnostic Systems.

Peptone Bacteriological Neutralized

A mixed pancreatic and papaic digest of animal tissues. It contains total nitrogen of 14.0% and NaCl of 1.6%. Available from BD Diagnostic Systems and Oxoid.

Peptone P

A peptic digest of fresh meat that has a high sulfur content and contains total nitrogen of 11.12% and NaCl of 9.3%. Available from BD Diagnostic Systems and Oxoid.

Peptonized Milk

A pancreatic digest of high grade skim milk powder. It has a high carbohydrate and calcium concentration. It contains total nitrogen of 5.3% and NaCl of 1.6%. Available from Oxoid.

Phytone™ Peptone

A papaic digest of soybean meal. It has a high vitamin and a high carbohydrate content. Available from BD Diagnostic Systems.

Polypeptone™ Peptone

A mixture of peptones composed of equal parts of pancreatic digest of casein and peptic digest of animal tissue. Available from BD Diagnostic Systems.

Proteose Peptone

A specialized peptone prepared from a mixture of peptones that contains a wide variety of high molecular weight peptides. It contains total nitrogen of 12.7% and NaCl of 8.0%. Available from BD Diagnostic Systems and Oxoid.

Proteose Peptone No. 2

An enzymatic digest of animal tissues with a high concentration of high molecular weight peptones. Available from BD Diagnostic Systems.

Proteose Peptone No. 3

An enzymatic digest of animal tissues. It has a high concentration of high molecular weight peptones. Available from BD Diagnostic Systems.

Soya Peptone

A papaic digest of soybean meal with a high carbohydrate concentration. It contains total nitrogen of 8.7% and NaCl of 0.4%. Available from Oxoid.

Soytone

A papaic digest of soybean meal. Available from BD Diagnostic Systems and Oxoid.

Special Peptone

A mixture of peptones, including meat, plant and yeast digests. It contains a wide variety of peptides, nucleotides, and minerals. It contains total nitrogen of 11.7% and NaCl of 3.5%. Available from Oxoid.

Thiotone™ E Peptone

An enzymatic digest of animal tissue. Available from BD Diagnostic Systems.

Trypticase™ Peptone

A pancreatic digest of casein. It has a very low carbohydrate content and a relatively high tryptophan content. Available from BD Diagnostic Systems.

Tryptone

A pancreatic digest of casein. It contains total nitrogen of 12.7% and NaCl of 0.4%. Available from Oxoid.

Tryptone T

A pancreatic digest of casein with lower levels of calcium, magnesium, and iron than tryptone. It contains total nitrogen of 11.7% and NaCl of 4.9%. Available from BD Diagnostic Systems and Oxoid.

Tryptose

An enzymatic hydrolysate containing high molecular weight peptides. It contains total nitrogen of 12.2% and NaCl of 5.7%. Available from BD Diagnostic Systems and Oxoid.

Meat and Plant Extracts

Meat and plant infusions are aqueous extracts that are commonly used as sources of nutrients for the cultivation of microorganisms. Such infusions contain amino acids and low molecular weight peptides, carbohydrates, vitamins, minerals, and trace metals.

Extracts of animal tissues contain relatively high concentrations of water soluble protein components and glycogen. Extracts of plant tissues contain relatively high concentrations of carbohydrates.

Below is a list of some of the meat and plant extracts that are used as ingredients in various media.

Bacto Beef

A desiccated powder of lean beef. Available from BD Diagnostic Systems.

Bacto Beef Extract

An extract of beef (paste). Available from BD Diagnostic Systems.

Bacto Beef Extract Desiccated

An extract of desiccated beef. Available from BD Diagnostic Systems.

Bacto Beef Heart for Infusion

A desiccated powder of beef heart. Available from BD Diagnostic Systems.

Bacto Liver

A desiccated powder of beef liver. Available from BD Diagnostic Systems.

Lab-Lemco

A meat extract powder. Available from Oxoid.

Liver Desiccated

Dehydrated ox livers. Available from Oxoid.

Malt Extract

A water soluble extract from germinated grain dried by low temperature evaporation. It has a high carbohydrate content. It contains total nitrogen of 1.1% and NaCl of 0.1%.

Growth Factors

Many microorganisms have specific growth factor requirements that must be included in media for their successful cultivation. Vitamins, amino acids, fatty acids, trace metals, and blood components often must be added to media. In some cases specific defined components are used to meet the growth factor requirements. Incorporation of growth factors are used to enrich, that is, to increase the numbers of particular species of microorganisms. Most often mixtures of growth factors are used in microbiological media. Acid hydrolysates of casein commonly are used as sources of amino acids. Extracts of yeast cells also are employed as sources of amino acids and vitamins for the cultivation of microorganisms. Many media, particularly those employed in the clinical laboratory, contain blood or blood components that serve as essential nutrients for fastidious microorganisms. X factor (heme) and V factor (nicotinamide adenine dinucleotide) often are supplied by adding hemoglobin (BD Diagnostic Systems), Iso-VitaleX (BD Diagnostic Systems), and/or Supple-

ment VX (BD Diagnostic Systems). Below is a list of some of the growth factors that are used as ingredients in various media.

Bacto Casamino Acids

A mixture of amino acids formed by acid hydrolysis of casein. Available from BD Diagnostic Systems.

Bacto Vitamin Free Casamino Acids

A mixture of amino acids formed by acid hydrolysis of casein that is free of vitamins. Available from BD Diagnostic Systems.

Bovine Albumin

Bovine albumin fraction V 0.2% in 0.85% saline solution. Available from BD Diagnostic Systems.

Bovine Blood, Citrated

Calf blood washed and treated with sodium citrate as an anticoagulant. Available from BD Diagnostic Systems.

Bovine Blood, Defibrinated

Calf blood treated to denature fibrinogen without causing cell lysis. Available from BD Diagnostic Systems.

Campylobacter Growth Supplement

Sodium pyruvate, sodium metabisulfite, and FeSO_4 .

CVA Enrichment

Glucose, L-cysteine-HCl·H₂O, vitamin B₁₂, L-glutamine, L-cystine-2HCl, adenine, nicotinamide adenine dinucleotide, cocarboxylase, guanine-HCl, $\text{Fe}(\text{NO}_3)_3$, *p*-aminobenzoic acid, and thiamine-HCl.

Cysteine Sulfide Reducing Agent

L-Cysteine-HCl·H₂O and Na₂S·9H₂O.

Dubos Medium Albumin

Albumin fraction V, glucose, and saline solution. Available from BD Diagnostic Systems.

Dubos Oleic Albumin Complex

Alkalinized oleic acid, albumin fraction V, and saline solution. Available from BD Diagnostic Systems.

Egg Yolk Emulsion

Chicken egg yolks and whole chicken egg. Available from BD Diagnostic Systems and Oxoid.

Egg Yolk Emulsion, 50%

Chicken egg yolks, whole chicken egg, and saline solution. Available from BD Diagnostic Systems.

EY Tellurite Enrichment

Egg yolk suspension with potassium tellu-

rite. Available from BD Diagnostic Systems and Oxoid.

Fresh Yeast Extract Solution

Live, pressed, starch-free, hydrolyzed baker's yeast.

Fildes Enrichment

A peptic digest of sheep or horse blood that is a rich source of growth factors including hemin and nicotinamide adenine dinucleotide. Available from BD Diagnostic Systems and Oxoid.

Hemoglobin

Dried bovine hemoglobin. Used to provide hemin required by many fastidious microorganisms. Available from BD Diagnostic Systems.

Hoagland Trace Element Solution, Modified

H₃BO₃, MnCl₂·4H₂O, AlCl₃, CoCl₂, CuCl₂, KI, NiCl₂, ZnCl₂, BaCl₂, Na₂MoO₄, SeCl₄, SnCl₂·2H₂O, NaVO₃·H₂O, KBr, and LiCl.

IsoVitaleX Enrichment

Glucose, L-cysteine-HCl, L-glutamine, L-cystine, adenine, nicotinamide adenine dinucleotide, vitamin B₁₂, thiamine pyrophosphate, guanine-HCl, $\text{Fe}(\text{NO}_3)_3$ ·6H₂O, *p*-aminobenzoic acid, and thiamine-HCl. Available from BD Diagnostic Systems.

Legionella Agar Enrichment

L-Cysteine and ferric pyrophosphate. Available from BD Diagnostic Systems.

Legionella BCYE Growth Supplement

ACES buffer/KOH, ferric pyrophosphate, L-cysteine-HCl, and α -ketoglutarate. For the enrichment of *Legionella* species. Available from Oxoid.

Leptospira Enrichment

Lyophilized pooled rabbit serum containing hemoglobin that provides long chain fatty acids and B vitamins for growth of *Leptospira* species. Available from BD Diagnostic Systems.

Metals "44"

ZnSO₄·7H₂O, FeSO₄·7H₂O, MnSO₄·7H₂O, CuSO₄·5H₂O, Co(NO₃)₂·6H₂O, EDTA, and Na₂B₄O₇·10H₂O.

Middlebrook ADC Enrichment

NaCl, bovine albumin fraction V, glucose, and catalase. The albumin binds free fatty acids that may be toxic to mycobacteria. Available from BD Diagnostic Systems.

Middlebrook OADC Enrichment

NaCl, bovine albumin, glucose, oleic acid, and catalase. The albumin binds free fatty acids that may be toxic to mycobacteria; the enrichment provides oleic acid used by *Mycobacterium tuberculosis* for growth. Available from BD Diagnostic Systems.

***Mycoplasma* Enrichment without Penicillin**

Horse serum, fresh autolysate of yeast—yeast extract, and thallium acetate. Provides cholesterol and nucleic acids for growth of *Mycoplasma* species. The thallium selectively inhibits other microorganisms. Available from BD Diagnostic Systems.

***Mycoplasma* Supplement**

Yeast extract and horse serum. Available from BD Diagnostic Systems.

Oleic Albumin Complex

NaCl, bovine albumin fraction V, and oleic acid. The albumin binds free fatty acids that may be toxic to mycobacteria and the enrichment provides oleic acid that is used by *Mycobacterium tuberculosis* for growth. Available from BD Diagnostic Systems.

RPF Supplement

Fibrinogen, rabbit plasma, trypsin inhibitor, and potassium tellurite. For the selection and nutrient supplementation of *Staphylococcus aureus*. Available from Oxoid.

SLA Trace Elements

$\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$, H_3BO_3 , $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, ZnCl_2 , $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$, $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, and $\text{Na}_2\text{SeO}_3 \cdot 5\text{H}_2\text{O}$.

Supplement A

Yeast concentrate with Crystal Violet. Available from BD Diagnostic Systems.

Supplement B

Yeast concentrate, glutamine, coenzyme, cocarboxylase, hematin, and growth factors. Available from BD Diagnostic Systems.

Supplement C

Yeast concentrate. Available from BD Diagnostic Systems.

Supplement VX

Essential growth factors V and X. Available from BD Diagnostic Systems.

Trace Element Mixture

Ethylenediamine tetraacetic acid (EDTA), $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, CaCl_2 , $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, and $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$.

Trace Element Solution HO-LE

H_3BO_3 , $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, sodium tartrate, $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$, $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, and ZnCl_2 .

Trace Elements Solution SL-6

H_3BO_3 , $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{Na}_2\text{MoO}_4 \cdot \text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, and $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$.

Trace Elements Solution SL-7

$\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, ZnCl_2 , H_3BO_3 , $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$, and HCl.

Trace Element Solution SL-8

Disodium EDTA, $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, ZnCl_2 , H_3BO_3 , $\text{NaMoO}_4 \cdot 2\text{H}_2\text{O}$, and $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$.

Trace Elements Solution SL-10

$\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, ZnCl_2 , $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$, H_3BO_3 , $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$ and HCl (25% solution).

Trace Metals A-5 Mix

H_3BO_3 , $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$, and $\text{Co}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$.

VA Vitamin Solution

Nicotinamide, thiamine-HCl, *p*-aminobenzoic acid, biotin, calcium pantothenate, pyridoxine-2HCl, and cyanocobalamin.

Vitamin K₁ Solution

Vitamin K₁ and ethanol.

Vitox Supplement

Glucose, L-cysteine-HCl, L-glutamine, L-cystine, adenine sulfate, nicotinamide adenine dinucleotide, cocarboxylase, guanine-HCl, $\text{Fe}(\text{NO}_3)_3 \cdot 6\text{H}_2\text{O}$, *p*-aminobenzoic acid, vitamin B₁₂, and thiamine-HCl. Available from Oxoid.

Yeast Autolysate Growth Supplement

Yeast autolysate fractions, glucose, and NaHCO_3 . Available from Oxoid.

Yeast Dialysate

Active, dried yeast.

Yeast Extract

A water soluble extract of autolyzed yeast cells. Available from BD Diagnostic Systems, and Oxoid.

Yeastolate

A water soluble fraction of autolyzed yeast cells rich in vitamin B complex. Available from BD Diagnostic Systems.

Selective Components

Many media contain selective components that inhibit the growth of nontarget microorganisms and favor the growth of specific organisms. Selective media are especially useful in the isolation of specific microorganisms from mixed populations. In many media for the study of microorganisms in nature, compounds are included in the media as sole sources of carbon or nitrogen so that only a few types of microorganisms can grow. Selective toxic compounds are also frequently used to select for the cultivation of particular microbial species. The isolation of a pathogen from a stool specimen, for example, where there is a high abundance of non-pathogenic normal microbiota, requires selective media. Often antimicrobics or other selectively toxic compounds are incorporated into media to suppress the growth of the background microbiota while permitting the cultivation of the target organism of interest. Bile salts, selenite, tetrathionate, tellurite, azide, phenylethanol, sodium lauryl sulfate, high sodium chloride concentrations, and various dyes—such as eosin, Crystal Violet, and Methylene Blue—are used as selective toxic chemicals. Antimicrobial agents used to suppress specific types of microorganisms include ampicillin, chloramphenicol, colistin, cycloheximide, gentamicin, kanamycin, nalidixic acid, sulfadiazine, and vancomycin. Various combinations of antimicrobics are effective in suppressing classes of microorganisms, such as enteric bacteria. Below are some of the selective agents, principally antimicrobial mixtures used for the selective isolation of pathogens, that are commonly used as selective agents in microbiological media.

Ampicillin Selective Supplement

Ampicillin. Used in media for the selection of *Aeromonas hydrophila*. Available from Oxoid.

Anaerobe Selective Supplement GN

Hemin, menadione, sodium succinate, nalidixic acid, and vancomycin. For the selection of Gram-negative anaerobes. Available from Oxoid.

Anaerobe Selective Supplement NS

Hemin, menadione, sodium pyruvate, and nalidixic acid. For the selection of non-sporulating anaerobes. Available from Oxoid.

Bacillus cereus Selective Supplement

Polymyxin B. For the selection of *Bacillus cereus*. Available from Oxoid.

Brucella Selective Supplement

Polymyxin B, bacitracin, cycloheximide, nalidixic acid, nystatin, and vancomycin. For the selection of *Brucella* species. Available from Oxoid.

Campylobacter Selective Supplement Blaser-Wang
Vancomycin, polymyxin B, trimethoprim, amphotericin B, cephalothin. For the selection of *Campylobacter* species. Available from Oxoid.

Campylobacter Selective Supplement Butzler
Bacitracin, cycloheximide, colistin sulfate, sodium cephalazolin, and novobiocin. For the selection of *Campylobacter* species. Available from Oxoid.

Campylobacter Selective Supplement Preston
Polymyxin B, rifampicin, trimethoprim, and cycloheximide. For the selection of *Campylobacter* species. Available from Oxoid.

Campylobacter Selective Supplement Skirrow
Vancomycin, trimethoprim, and polymyxin B. For the selection of *Campylobacter* species. Available from Oxoid.

CCDA Selective Supplement

Cefoperazone and amphotericin B. For the selection of *Campylobacter* species. Available from Oxoid.

Cefoperazone Selective Supplement

Cefoperazone. For the selection of *Campylobacter* species. Available from Oxoid.

Chapman Tellurite Solution

Potassium tellurite 1% solution. Available from BD Diagnostic Systems.

Clostridium difficile Selective Supplement

D-Cycloserine and cefoxitin. For the selection of *Clostridium difficile*. Available from BD Diagnostic Systems and Oxoid.

Fraser Supplement

Ferric ammonium sulfate, nalidixic acid, and Acriflavin hydrochloride. For the selection of *Listeria* species. Available from Oxoid.

Kanamycin Sulfate Selective Supplement

Kanamycin sulfate. For the selection of enterococci. Available from Oxoid.

Listeria Primary Selective Enrichment Supplement

Nalidixic acid and acriflavin. For the selection of *Listeria* species. Available from Oxoid.

Listeria Selective Enrichment Supplement

Nalidixic acid, cycloheximide, and acriflavin. For the selection of *Listeria* species. Available from Oxoid.

Listeria Selective Supplement MOX

Colistin and moxalactam. For the selection of *Listeria monocytogenes*. Available from Oxoid.

Listeria Selective Supplement Oxford

Cycloheximide, colistin sulfate, acriflavin, cefotetan, and fosfomycin. For the selection of *Listeria* species. Available from Oxoid.

Modified Oxford Antimicrobial Supplement

Moxalactam and colistin sulfate. Available from BD Diagnostic Systems.

Oxgall

Dehydrated fresh bile. For the selection of bile tolerant bacteria. Available from BD Diagnostic Systems.

Oxytetracycline GYE Supplement

Oxytetracycline in a buffer. For the selection of yeasts and filamentous fungi. Available from Oxoid.

PALCAM Selective Supplement

Polymyxin B, acriflavin hydrochloride, and ceftazidime. For the selection of *Listeria monocytogenes*. Available from Oxoid.

Perfringens OPSP Selective Supplement A

Sodium sulfadiazine. For the selection of *Clostridium perfringens*. Available from Oxoid.

Perfringens SFP Selective Supplement A

Kanamycin sulfate and polymyxin B. For the selection of *Clostridium perfringens*. Available from Oxoid.

Perfringens TSC Selective Supplement A

D-Cycloserine. For selection of *Clostridium perfringens*. Available from Oxoid.

Sodium Desoxycholate

Sodium salt of desoxycholic acid. Available from BD Diagnostic Systems.

Staph/Strep Selective Supplement

Nalidixic acid and colistin sulfate. For the selection of *Staphylococcus* species and *Streptococcus* species. Available from Oxoid.

***Streptococcus* Selective Supplement COA**

Colistin sulfate and oxolinic acid. For the selection of *Streptococcus* species. Available from Oxoid.

Sulfamandelate Supplement

Sodium sulfacetamide and sodium mandelate. For the selection of *Salmonella* species. Available from Oxoid.

Tellurite Solution

A solution containing potassium tellurite. Inhibits Gram-negative and most Gram-positive microorganisms. It is used for the isolation of *Corynebacterium* species, *Streptococcus* species, *Listeria* species, and *Candida albicans*. Available from BD Diagnostic Systems.

***Yersinia* Selective Supplement**

Cefsulodin, irgasan, and novobiocin. For the selection of *Yersinia enterocolitica*. Available from Oxoid.

pH Buffers

Phosphate buffers commonly are used to maintain the pH of media. The pH is established by using varying volumes of equimolar concentrations of Na_2HPO_4 and NaH_2PO_4 .

pH	Na_2HPO_4 (mL)	NaH_2PO_4 (mL)
5.4	3.0	97.0
5.6	5.0	95.0
5.8	7.8	92.2
6.0	12.0	88.0
6.2	18.5	81.5
6.4	26.5	73.5
6.6	37.5	62.5
6.8	50.0	50.0
7.0	61.1	38.9
7.2	71.5	28.5
7.4	80.4	19.6
7.6	86.8	13.2
7.8	91.4	8.6
8.0	94.5	5.5

Differential Components

The differentiation of many microorganisms is based upon the production of acid from various carbohydrates and other carbon sources or the decarboxylation of amino acids. Some media include indicators, particularly of pH that permit the visual detection of changes in pH resulting from such metabolic reactions. Below is a list of some commonly used pH indicators and their color reactions.

pH Indicator	pH Range	Acid Color	Alkaline Color
<i>m</i> -Cresol Purple	0.5–2.5	Red	Yellow
Thymol Blue	1.2–2.8	Red	Yellow
Bromphenol Blue	3.0–4.6	Yellow	Blue
Bromcresol Green	3.8–5.4	Yellow	Blue
Chlorcresol Green	4.0–5.6	Yellow	Blue
Methyl Red	4.2–6.3	Red	Yellow
Chlorphenol Red	5.0–6.6	Yellow	Red
Bromcresol Purple	5.2–6.8	Yellow	Purple
Bromthymol Blue	6.0–7.6	Yellow	Blue
Phenol Red	6.8–8.4	Yellow	Red
Cresol Red	7.2–8.8	Yellow	Red
<i>m</i> -Cresol Purple	7.4–9.0	Yellow	Purple
Thymol Blue	8.0–9.6	Yellow	Blue
Cresolphthalein	8.2–9.8	Colorless	Red
Phenolphthalein	8.3–10.0	Colorless	Red

Many new media for the rapid detection of microorganisms in foods utilize chromogenic and fluorogenic indicators. Chromogenic and fluorogenic substrates consist of a chromophor or fluorophor,

respectively, that is linked to a substrate for a specific diagnostic enzyme. The enzyme releases the chromophor or fluorophor which can then easily be recognized by its characteristic color. Below is a list of some of the chromogenic and fluorogenic indicators and substrates that are commonly used in media for the detection of specific microorganisms.

Chromophor/Fluorophor	Color
5-Bromo-4-chloro-3-indoxyl	Blue-truquoise/ mint-green
5-Bromo-3-indoxyl	Blue
5-Bromo-6-chloro-3-indoxyl	Purple/ magenta
6-Chloro-3-indoxyl	Rose/salmon
N-Methylindoxyl-	Green
2-Nitrophenyl	Yellow
4-Methylumbelliferyl-	Blue (UV 365nm)
Cresolphthalein	Red
Substrate	Target Microbes
–β-D-glucuronide	<i>E. coli</i>
–β-D-galacto-pyranoside	Coliforms, Enterobacteriaceae
–β-D-gluco-pyranoside	<i>Enterococcus</i> spp.
–α-D-gluco-pyranoside	<i>Enterococcus</i> spp.
–caprylate-nonanoate	<i>Salmonella</i> spp.
–myo-inositol-1-phosphate	<i>Listeria</i> spp. <i>B. cereus</i>
–phosphate	<i>S. aureus</i> <i>C. perfringens</i>
–N-acetyl-β-D-	<i>Candida</i> spp.
–N-acetyl-β-D-galac- tosaminide	<i>Candida</i> spp.

Preparation of Media

The ingredients in a medium are usually dissolved and the medium is then sterilized. When agar is used as a solidifying agent the medium must be heated gently, usually to boiling, to dissolve the agar. In some cases where interactions of components, such as metals, would cause precipitates, solutions must be prepared and occasionally sterilized separately before mixing the various solutions to prepare the complete medium. The pH often is adjusted prior to sterilization, but in some cases sterile acid or base is used to adjust the pH of the medium following sterilization. Many media are sterilized by exposure to elevated temperatures. The most common method is to autoclave the medium. Different sterilization procedures are employed when heat-labile compounds are included in the formulation of the medium.

Autoclaving

Autoclaving uses exposure to steam, generally under pressure, to kill microorganisms. Exposure for 15 min to steam at 15 psi—121°C is most commonly used. Such exposure kills vegetative bacterial cells and bacterial endospores. However, some substances do not tolerate such exposures and lower temperatures and different exposure times are sometimes employed. Media containing carbohydrates often are sterilized at 116°C to 118°C in order to prevent the decomposition of the carbohydrate and the formation of toxic compounds that would inhibit microbial growth.

Below is a list of pressure–temperature relationships.

Pressure (psi)	Temperature (°C)
0	100
1	101.9
2	103.6
3	105.3
4	106.9
5	108.4
6	109.8
7	111.3
8	112.6
9	113.9
10	115.2
11	116.4
12	117.6
13	118.8
14	119.9
15	121.0
16	122.0
17	123.0
18	124.0
19	125.0
20	126.0
21	126.9
22	127.8
23	128.7
24	129.6
25	130.4

Caution about Hazardous Components

Some media contain components that are toxic or carcinogenic. Appropriate safety precautions must be taken when using media with such components. Basic fuchsin and acid fuchsin are carcinogens and caution must be used in handling media with these compounds to avoid dangerous exposure that could lead to the development of malignancies. Thallium salts, sodium azide, sodium biselenite, and cyanide are among the toxic components found in some media. These compounds are poisonous and steps

must be taken to avoid ingestion, inhalation, and skin contact. Azides also react with many metals, especially copper, to form explosive metal azides. The disposal of azides must avoid contact with copper or achieve sufficient dilution to avoid the formation of such hazardous explosive compounds. Media with sulfur-containing compounds may result in the formation of hydrogen sulfide which is a toxic gas. Care must be used to ensure proper ventilation. Media with human blood or human blood components must be handled with great caution to avoid exposure to human immunodeficiency virus and other pathogens that contaminate some blood supplies. Proper handling and disposal procedures must be followed with blood-containing as well as other media that are used to cultivate microorganisms.

FDA Recommended Media for the Examination of Foods

Below is a list of the FDA Recommended Media for the Testing of Foods listed in the *Bacteriological Analytical Manual*:

- M1 A-1 Medium
- M2 Acetamide Medium
- M3 Acetate Agar
- M4 Acid Broth
- M5 AE Sporulation Medium, Modified (for *C. perfringens*)
- M6 Agar Medium P
- M7 AKI Medium
- M8 Alkaline Peptone Agar
- M9 Alkaline Peptone Salt Broth (APS)
- M10 Alkaline Peptone Water
- M10a ALOA (Agar *Listeria* Ottavani & Agosti) Medium
- M11 Anaerobe Agar
- M12 Anaerobic Egg Yolk Agar
- M13 Andrade's Carbohydrate Broth and Indicator
- M14 Antibiotic Medium No. 1 (Agar Medium A)
- M15 Antibiotic Medium No. 4 (Agar Medium B)
- M16 Arginine-Glucose Slant (AGS)
- M17 Baird-Parker Medium, pH 7.0
- M18 Bile Esculin Agar
- M17a Biosynth Chromogenic Medium (BCM) for *Listeria monocytogenes*
- M19 Bismuth Sulfite Agar (Wilson and Blair)
- M20 Blood Agar
- M20a Blood Agar Base
- M21 Blood Agar Base (Infusion Agar)
- M22 Blood Agar Base #2 (Difco)
- M23 Brain Heart Infusion (BHI) Agar (0.7%) (for staphylococcal enterotoxin)
- M24 Brain Heart Infusion (BHI) Broth and Agar
- M25 Brilliant Green Lactose Bile Broth
- M26 Bromcresol Purple Broth
- M27 Bromcresol Purple Dextrose Broth (BCP)
- M28a *Campylobacter* enrichment broth (Bolton formula)
- M29a Abeyta-Hunt-Bark Agar
- M30a Modified *Campylobacter* Blood-Free Selective Agar Base (CCDA)
- M30b Freezing medium.
- M30c Semi-Solid Medium, modified, for Culture Storage
- M30d Semisolid Medium, modified, for Biochemical Identification
- M31 Cary-Blair transport Medium
- M32 Casamino Acids-Yeast Extract (CYE) Broth
- M34 Casamino Acids-Yeast Extract-Salts (CA-YE) Broth (Gorbach)
- M35 Cefsulodin-Irgasan Novobiocin (CIN) Agar
- M35 *Yersinia* Selective Agar (YSA)
- M36 Cell Growth Medium
- M37 Cetrimide Agar
- M38 Chopped Liver Broth
- M39 Christensen Citrate Agar
- M40 Christensen's Urea Agar
- M40a CHROMagar *Listeria*
- M40b Chromogenic *Listeria* Agar
- M41 Congo Red BHI Agarose (CRBHO) Medium
- M42 Cooked Meat Medium
- M43 Cooked Meat Medium (Modified)
- M44 Decarboxylase Basal Medium (Arginine, Lysine, Ornithine)
- M45 Duncan-Strong (DS) Sporulation Medium, Modified (for *C. perfringens*)
- M46 Eagle's Minimal Essential Medium (MEME) (with Earle's salts and nonessential amino acids)
- M47 Earle's Balanced Salts (Phenol Red-Free)
- M48 EB Motility Medium
- M49 EC Broth
- M50 EC-MUG Medium
- M51 Egg Yolk Emulsion, 50%
- M52 Buffered *Listeria* Enrichment Broth, pH 7.3 ± 0.1
- M53 Esculin Agar, Modified (CDC)
- M54 Gelatin Agar (GA)
- M55 Gelatin Salt Agar (GS)
- M56a Papain Solution, 5%
- M57 Gentamicin Sulfate Solution
- M58 Ham's F-10 Medium
- M59 Heart infusion agar (HIA) (Difco)
- M60 Heart Infusion (HI) Broth and Agar (HIA) (for *Vibrio*)
- M61 Hektoen Enteric (HE) Agar
- M62 HC (Hemorrhagic colitis *E. coli* strains) Agar
- M63 Hugh-Leifson Glucose Broth (HLGB)
- M64 Indole Medium
- M65 Indole Medium (CDC)

- M66 Indole Nitrite Medium (tryptic Nitrate)
 M67 Irgasan-Ticarcillin-Chlorate (ITC) Broth
 M68 Iron Milk Medium (Modified)
 M69 King's B Medium
 M70 King's O/F Basal Medium
 M71 Kligler Iron Agar
 M72 Koser's Citrate Broth
 M73 L-15 Medium (Modified) Leibovitz
 M74 Lactose Broth
 M75 Lactose-Gelatin Medium (for *C. perfringens*)
 M76 Lauryl tryptose (LST) Broth
 M77 Lauryl tryptose MUG (LST-MUG) Broth
 M78 Lethen Agar (Modified)
 M79 Lethen Broth (Modified)
 M80 Levine's Eosin-Methylene Blue (L-EMB) Agar
 M81 Lithium Chloride-Phenylethanol-Moxalactam (LPM) Medium
 M82 [Lithium Chloride-Phenylethanol-Moxalactam] LPM Plus Esculin and Ferric Iron
 M83 Liver-Veal Agar
 M84 Liver-Veal-Egg Yolk Agar
 M85 Long-term Preservation Medium
 M86 Lysine Arginine Iron Agar (LAIA)
 M87 Lysine Decarboxylase Broth (Falkow) (for *Salmonella*)
 M88 Lysine Decarboxylase (LDC) Medium (for Gram-negative nonfermentative bacteria)
 M89 Lysine Iron Agar (Edwards and Fife)
 M90 Lysozyme Broth
 M91 MacConkey Agar
 M92 Malonate Broth
 M93 Malt Extract Agar (Cosmetics-General Microbiology)
 M94 Malt Extract Broth (Difco)
 M95 Mannitol-Egg Yolk-Polymyxin (MYP) Agar
 M96 Mannitol Maltose Agar
 M97 Mannitol Salt Agar
 M98 Modified Cellobiose-Polymyxin B-Colistin (mCPC) Agar
 M103a Modified Oxford *Listeria* Selective Agar
 M99 Motility-Indole-Ornithine (MIO) Medium
 M100 Motility Medium (for *B. cereus*)
 M101 Motility Nitrate Medium (for Cosmetics)
 M102 Motility-Nitrate Medium, Buffered (for *C. perfringens*)
 M103 Motility Test Medium (Semisolid)
 M104 MR-VP Broth
 M105 Mucate Broth
 M106 Mucate Control Broth
 M107 Mueller-Hinton Agar
 M108 Nitrate Broth
 M109 Nitrate Broth, Enriched (CDC)
 M110 Nitrate Reduction Medium and Reagents
 M111 Nonfat Dry Milk (Reconstituted)
 M112 Nutrient Agar
 M113 Nutrient Agar (for *B. cereus*)
 M114 Nutrient Broth
 M115 Nutrient Gelatin (CDC) (for Gram-negative nonfermentative bacteria)
 M116 OF Glucose Medium, Semisolid
 M117 Oxidative-Fermentative (OF) Test Medium
 M118 Oxford Medium
 M118a PALCAM *Listeria* Selective Agar
 M119 Penicillin-Streptomycin Solution
 M120 Peptone Sorbitol Bile Broth
 M121 Phenol Red Carbohydrate Broth
 M122 Phenol Red Glucose Broth
 M123 Phenylalanine Deaminase Agar
 M124 Plate Count Agar (Standard Methods)
 M125 PMP Broth
 M126 Potassium Cyanide (KCN) Broth
 M127 Potato Dextrose Agar
 M128 *Pseudomonas* Agar F (for fluorescein production)
 M129 *Pseudomonas* Agar P (for pyocyanine production)
 M130 Purple Carbohydrate Broth
 M130a Purple Carbohydrate Fermentation Broth Base
 M131 Pyrazinamidase Agar*
 M131a Rapid *L. mono* Medium
 M132 Rappaport-Vassiliadis Medium
 M133 Sabouraud's Dextrose Broth and Agar
 M134 Selenite Cystine Broth
 M135 Sheep Blood Agar
 M136 *Shigella* Broth
 M137 SIM Motility Medium
 M138 Simmons Citrate Agar
 M139 Sorbitol-MacConkey Agar
 M140 Sporulation Broth (for *C. perfringens*)
 M141 Spray's Fermentation Medium (for *C. perfringens*)
 M142 *Staphylococcus* Agar No. 110
 M143 Starch Agar
 M144 T₁N₁ Medium
 M145 Tetrathionate Broth
 M146 Thioglycollate Medium (Fluid) (FTG)
 M147 Thiosulfate-Citrate-Bile Salts-Sucrose (TCBS) Agar
 M148 Toluidine Blue-DNA Agar
 M149 Triple Sugar Iron Agar (TSI)
 M150 Trypticase Novobiocin (TN) Broth
 M151 Trypticase-Peptone-Glucose-Yeast Extract Broth (TPGY)
 M151a Trypticase-Peptone-Glucose-Yeast Extract Broth with Trypsin (TPGYT)
 M152 Trypticase (Tryptic) Soy Agar
 M152a Trypticase Soy Agar-Magnesium sulfate-NaCl (TSAMS)

M153 Trypticase Soy Agar with 0.6% Yeast Extract (TSAYE)
 M154 Trypticase (Tryptic) Soy Broth
 M154a Trypticase (Tryptic) Soy Broth with 10% NaCl and 1% Sodium Pyruvate
 M155 Trypticase (Tryptic) Soy Broth (TSB) with Glycerol
 M156 Trypticase Soy Broth Modified (mTSB)
 M157 Trypticase Soy Broth with 0.6% Yeast Extract (TSBYE)
 M158 Trypticase Soy-Polymyxin Broth
 M159 Trypticase Soy-Sheep Blood Agar
 M160 Trypticase Soy-Tryptose Broth
 M161 Tryptone Broth and Tryptone Salt Broths
 M162 Tryptone Phosphate (TP) Broth
 M163 Tryptone Salt (T_1N_1) Agar and T_1N_2 Agar
 M164 Tryptone (Tryptophane) Broth, 1%
 M165 Tryptone Yeast Extract Agar
 M166 Tryptose Blood Agar Base
 M167 Tryptose Broth and Agar (for serology)
 M168 Tryptose Phosphate Broth (TPB)
 M169 Tryptose-Sulfite-Cycloserine (TSC) Agar
 M170 Tyrosine Agar
 M171 Urea Broth
 M172 Urea Broth (Rapid)
 M173 Veal Infusion Agar and Broth
 M174 Violet Red Bile Agar (VRBA)
 M175 Violet Red Bile-MUG Agar (VRBA-MUG)
 M176 Vogel-Johnson (VJ) Agar
 M177 Voges-Proskauer Medium (Modified)
 M178 Wagatsuma Agar
 M179 Xylose Lysine Desoxycholate (XLD) Agar
 M180 Y-1 Adrenal Cell Growth Medium
 M181 Yeast Extract (YE) Agar
 M182 Malt Extract Agar (Yeasts and Molds) (MEAYM)
 M183 Dichloran rose bengal chloramphenicol (DRBC) agar
 M184 Dichloran 18% glycerol (DG18) agar
 M185 Malt Agar (MA)

M186 Trypticase (Tryptic) Soy Broth (TSB) with Ferrous Sulfate

M187 Cellobiose-Colistin (CC) Agar

M188 Universal Preenrichment Broth

M189 Cellulase Solution

M190 *Vibrio vulnificus* Agar (VVA)

M191 *Vibrio parahaemolyticus* sucrose Agar (VPSA)

References

Below is a list of references that can be consulted for further information about media used for the isolation, cultivation, and differentiation of microorganisms from foods.

Compendium of Methods for the Microbiological Examination of Foods. 1992. C. Vanderzant and D. F. Splittstoesser, eds. American Public Health Association, Washington, D.C.

Difco™ & BBL™ Manual: Dehydrated Culture Media and Reagents for Microbiology. 2003. BD Diagnostic Systems, Sparks, MD. <http://www.bd.com/ds/technicalCenter/inserts/difcoBblManual.asp>

Food and Drug Administration Bacteriological Analytical Manual (BAM). 2001. U.S. Food and Drug Administration (FDA). <http://www.cfsan.fda.gov/~ebam/bam-toc.html>

Food Microbiology (07.100.30). International Organization for Standardization. <http://www.iso.org>

New developments in chromogenic and fluorogenic culture media. 2000. M. Manafi. International Journal of Food Microbiology 60:205–218

The Oxoid Manual. 1990. E.Y. Bridson, ed. Unipath Ltd. Basingstoke, Hampshire England.

Practical Food Microbiology. Third Edition. 2002. D. Roberts. Blackwell Synergy Publishing, Oxford England.

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