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Good hygienic practice in milk processing

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4.1 Introduction

Milk and dairy products are highly nutritious media, in which micro-organisms can multiply and cause spoilage. The levels and types of micro-organisms in milk and dairy products depend on the microbial quality of the raw materials, the conditions under which the products are produced and the temperature and duration of storage. The most common spoilage micro-organisms of milk and dairy products are Gram-negative rod-shaped bacteria (e.g. *Pseudomonas* spp., coliforms), Gram-positive spore-forming bacteria (e.g. *Bacillus* spp., *Clostridium* spp.), lactic acid producing bacteria (e.g. *Streptococcus* spp.) and yeasts and moulds.

Milk and milk products are also, to a limited extent, associated with foodborne illness. In the USA milk was involved in 0.2–0.5% of the foodborne disease outbreaks with known vehicle in the period 1993–1997, i.e. 3–105 reported cases per year. In the Netherlands, investigations of foodborne diseases in 1991–1994 showed that dairy products were involved in about 3% of reported cases with known etiological agent. More than 90% of all reported cases of dairy-related illness are of bacterial origin. Disease is mainly due to consumption of unpasteurized milk containing pathogenic micro-organisms (e.g. *Salmonella*, *Listeria monocytogenes* or *Campylobacter*).

In this chapter, the principal microbial hazards concerning milk and milk products, focused on heat-treated liquid milk products, will be described. The importance of good hygienic practice measures and dairy product safety systems at farm, processing plant and consumer level will be discussed. Finally, some future trends in dairy processing are presented.

4.2 The principal hazards

4.2.1 Raw milk

Raw milk, as secreted by healthy cows, is free of micro-organisms. However, micro-organisms associated with the teat move up the teat canal and into the interior of the udder. Most of the bacteria present in raw milk are contaminants of the outside and gain entrance into the milk from various sources including soil, bedding, manure, feed and milking equipment. Therefore, raw milk contains levels of a few to several thousands of bacteria. The microbial quality and the composition of the microflora of raw milk vary with seasons. Improvement of handling and processing of milk such as developments in closed milking systems, use of bulk tanks to store and transport raw milk and changes in refrigeration systems have resulted in shifts in the microflora from predominantly Gram-positive, acid-producing bacteria to Gram-negative, psychrotrophic micro-organisms, mainly *Pseudomonas* species. They grow rapidly at refrigeration temperatures and produce heat-resistant extracellular proteolytic and lipolytic enzymes that survive heat processing. Enzyme activity during storage will result in defects in flavour, texture and stability in milk and dairy products.

A variety of pathogenic bacteria have been isolated from raw milk including *Mycobacterium* spp., *Salmonella*, *Listeria monocytogenes*, *Bacillus cereus*, *Campylobacter jejuni*, *Yersinia enterocolitica*, *Escherichia coli* and *Staphylococcus aureus*. Depending on the country of origin, species, climate and sanitary conditions, raw milk can contain one or more of the pathogens listed.

4.2.2 Pasteurized milk

The health rules for the production and placing on the market of raw milk, heat-treated milk and milk-based products are described in Council Directive 92/46/EEC. Pasteurization is applied to destroy heat-sensitive spoilage and pathogenic bacteria present in the raw milk. The minimum requirements to destroy potential pathogenic micro-organisms are pasteurization for 15 s at 71.7°C or for 30 min at 62.7°C. In Fig. 4.1, the inactivation of vegetative (pathogenic) micro-organisms (i.e. *Listeria monocytogenes* and *Salmonella*) and bacterial spores (*Clostridium tyrobutyricum* and *Bacillus stearothermophilus*) by various heat treatments is shown. The figures demonstrate that the spores are much more resistant to heat than the vegetative micro-organisms. As can be observed, thermization and pasteurization are not sufficient to inactivate bacterial spores.

Further processing steps (pH reduction through the addition of starter, drying, addition of salt and cooling) are designed to limit the growth of the thermoduric bacteria that survive the heat treatment. Spoilage of pasteurized milk products is caused by:

- Growth and enzyme production by psychrotrophs before pasteurization
- Activity of thermostable enzymes

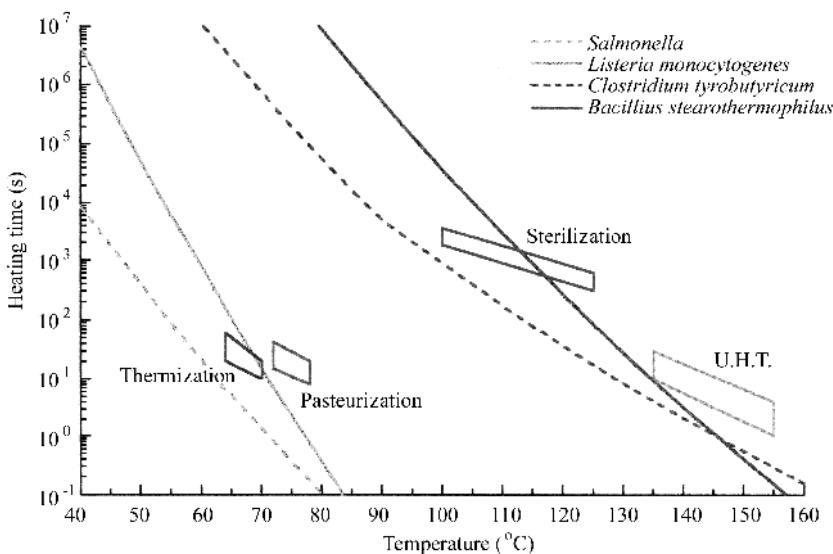


Fig. 4.1 Inactivation of micro-organisms in raw milk by various heat treatments applied in the processing of milk and dairy products.

- Growth of thermoresistant psychrotrophs
- Post-pasteurization contamination via equipment (pumps, valves, pipes, pasteurizers, storage tanks, filling equipment).

The effectiveness of cleaning and sanitizing procedures greatly influences the level of contamination and the types of micro-organisms introduced via equipment.

Trends in the dairy industry such as the extended refrigerated storage of raw milk prior to processing, the application of higher pasteurization temperatures and the more effective control of post-pasteurization contamination have enhanced the importance of thermoduric psychrotrophs. Spore-forming bacteria, predominantly *Bacillus* species, limit the shelf-life of pasteurized milk and milk products. Especially *B. cereus* is associated with defects such as off-flavours, sweet curdling and bitty cream caused by proteinase, lipase and phospholipases produced by the bacteria. Several studies have shown that *B. cereus* was present in pasteurized milk after storage.

In properly processed dairy products, most pathogens are not considered a problem, since pasteurization is effective in destroying these organisms. However, several cases of foodborne illness have been reported for, e.g., *Salmonella*, *Listeria*, *E. coli* and *Yersinia*, due to post-pasteurization contamination. Production of heat-stable enterotoxins by *St. aureus* in raw milk may also cause disease via various dairy products including pasteurized milk, cheese, ice cream, butter and non-fat dry milk.

4.2.3 UHT-milk

Sterilization is intended to destroy all the micro-organisms present, both vegetative forms and spores, or at least make them incapable of growth in the product, so that a long keeping quality is obtained without refrigerated storage.

The Milk Hygiene Directive 92/46/EEC demands that the minimum heating temperature for the manufacture of UHT-milk should be 135°C with a minimum holding time of 1 s. Typical time-temperature combinations applied in the dairy industry are holding times of the order of a few seconds at temperatures ranging from 135 to 150°C.

There are many different types of UHT-sterilizing equipment. The principles of operation and construction of the main types of equipment are summarized in Fig. 4.2.

Microbial spoilage of UHT-milk may occur by outgrowth of spores, surviving the heat processing, or by post-process contamination after heat processing (e.g. via packaging material or cooling water) or a failure in the thermal process. Typical spoilage organisms include thermoduric and spore-forming bacteria such as *Bacillus* species, *Streptococcus* and *Micrococcus* and occasionally some Gram-negative bacteria. Spores of *Bacillus* spp., e.g. *B. subtilis*, *B. megaterium*, *B. coagulans* and *B. stearothermophilus*, survive UHT-processing and can affect products such as canned or UHT-products. Defects include gas production, acid coagulation, thinning, bitterness and off-odours.

Since 1985, the occurrence of highly heat-resistant mesophilic spores (HRS) or *B. sporothermodurans*, causing non-sterility in UHT-sterilized dairy products, e.g. milk, chocolate milk, evaporated milk, reconstituted milk and cream, has been reported. The stability or sensoric quality is not altered by growth of these spore-formers. Coagulation and a pinkish colouration were only found after extended storage. All investigated *B. sporothermodurans* isolates show high resistance in the UHT-region, with D_{140} values ranging from 3.4 to 7.9 s, compared with *B. stearothermophilus* with a D_{140} value of 0.9 s. In the range 110–120°C, the spores of *B. sporothermodurans* are just as heat resistant (or less) as those of *B. stearothermophilus*. This is shown in Fig. 4.3.

Bacillus sporothermodurans is not a risk to the health of consumers. However, dairies are forced to manage the problem due to legal requirements

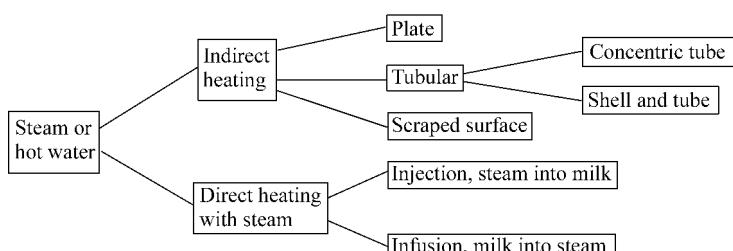


Fig. 4.2 Types of UHT-processing equipment

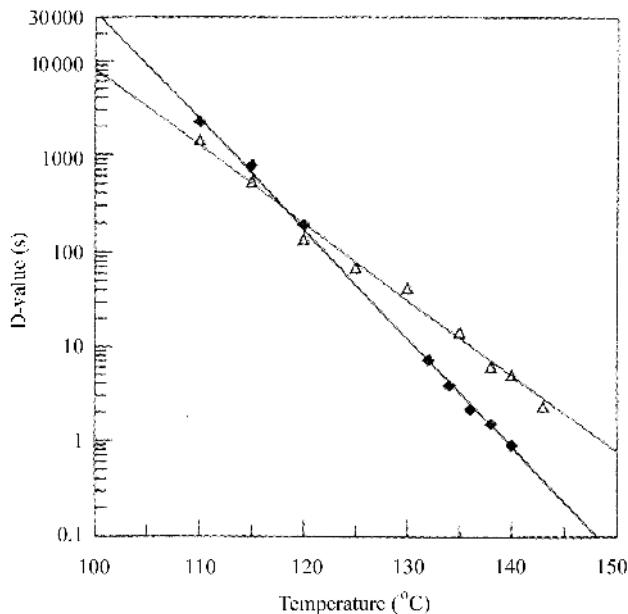


Fig. 4.3 Thermal death time curves of *B. stearothermophilus* spores (■) and *B. sporothermodurans* J16 (▲); best fit lines through experimental data.

and to avoid trade restrictions. To control *B. sporothermodurans*, direct or indirect/direct heating processes reaching F_0 values of >50 are necessary.

In addition to microbial spoilage of UHT-milk, gelation and coagulation of milk proteins and off-flavour formation may also occur as a result of heat-resistant proteolytic or lipolytic enzymes produced in the raw milk during storage. Proteolytic enzymes, naturally present in milk, probably originating from blood, are heat resistant. Studies have shown that these proteinases could survive UHT-processing.

4.3 Good hygienic practice

The various stages in the milk processing chain, from milking the cow to consumption, have to be under control in order to assure the quality and safety of milk and dairy products. Adherence to basic good manufacturing practices is one of the first steps to achieve this. Furthermore, HACCP can be applied as a tool to assess hazards and establish control systems that focus on preventive measures rather than relying mainly on end-product testing. Critical key aspects with respect to milk and dairy products are ensuring that raw materials are of the best quality, elimination of spoilage and pathogenic bacteria from raw milk and other raw materials by heat treatment, prevention of subsequent contamination, and growth limitation of undesirable micro-organisms during storage prior to consumption.

4.3.1 Farm

Micro-organisms and spores are widespread in the natural environment, with soil, water, plants and animals serving as reservoirs. Some degree of contamination of raw milk during production is inevitable, milking and milk storage equipment being the major sources of contamination. If milk is produced under sanitary conditions, the typical bacteria of the udder surface, mainly Micrococcaceae, predominate and less than 10% of the total flora are psychrotrophs. Under unsanitary conditions of production, milk can contain more than 75% psychrotrophs.

The occurrence of various bacterial species adhering to rubber and stainless steel in a milking installation has been reported. Gram-negative organisms predominated (96–100%), the majority being *Acinetobacter* spp., followed by *Pseudomonas* spp. and *Flavobacterium* spp.

The numbers and types of micro-organisms that develop subsequently during refrigerated storage are determined by the temperature and duration of the storage.

It is unlikely that all bacteria can be eliminated from the raw milk supply. Most important is to minimize contamination at the farm and keep the levels as low as possible by good hygienic practices. These include proper cleaning and sanitizing of milking equipment and rapid cooling to temperatures of 4°C or less.

The raw milk must be transported to the dairy under such conditions that the microbiological quality of the milk is not reduced. Milk collection tankers should be designed and constructed according to the IDF Code of Practice for Design and Construction of Milk Collection Tankers (IDF Document 128). During transport, the temperature of the milk should not exceed 7°C. Insulation and refrigeration of milk tankers may be necessary under some climatic conditions. The milk tanker should be cleaned and disinfected at least daily and whenever there is a gap of 4 hours or greater between collections. The sufficiency of cleaning and disinfection should be checked regularly.

4.3.2 Processing

At all stages in processing, good hygiene of the manufacturing plant is essential to ensure that the product stream is not (re)contaminated after heat treatment of raw milk (pasteurization or UHT-sterilization). Sources of post-pasteurization include equipment, packaging materials, air, aerosols, (condensed) water, lubricants, etc. Pasteurization equipment should be properly designed, installed, maintained and operated to ensure that the milk is heated to at least the specified temperature for at least the specified time.

Requirements for good hygiene design of food processing equipment, including dairy equipment, are described in various directives of the European Commission, the Hygiene of Foodstuffs' Directive and the Machinery Safety Directive (89/392/EEC). In addition, CEN/TC 153 has produced a European Standard on the hygienic requirements for food processing machinery to support 89/392/EEC.

Various organizations such as the European Hygienic Equipment Design Group (EHEDG), the International Dairy Federation (IDF), the 3-A organization and the International Standardization Organization (ISO, Technical Committee 199) have formulated and published (voluntary) principles of hygienic and aseptic design, requirements for hygienic and aseptic equipment and methods to test whether equipment fulfils these requirements. Guidance on design, construction and installation of equipment, cleaning-in-place (CIP) systems and plant is given in various IDF documents, e.g. IDF Docs 117, 123, 218, 292. Summaries of EHEDG guideline documents are published by Elsevier in *Trends in Food Science and Technology*. 3-A sanitary standards are available for many types of equipment, from fittings to silo tanks. Documents are published in *Dairy, Food and Environmental Sanitation (DFES)* magazine.

To maintain the factory environment in a hygienic condition, cleaning programmes should be established. Most of the equipment used for handling milk and milk products is cleaned and disinfected by CIP systems at least daily. Start-up of closed processing lines in the dairy industry is usually done by circulating hot water in order to have additional decontamination of the equipment. Monitoring CIP systems, i.e. concentrations of the cleaning agents, temperatures, flow, pressure and circulation time, is necessary to ensure the efficiency of cleaning.

Biofilms present on the surface of milk processing equipment threaten the quality and safety of dairy products. Dead ends, corners, cracks, crevices, gaskets, valves and joints are vulnerable points for biofilm accumulation. Development of biofilms in a dairy manufacturing plant depends on the type of micro-organism, the type of product being processed, the operating conditions of the plant (temperatures, length of production runs) and the type of surface. The hygienic design of processing equipment is of great importance in avoiding biofilm formation. Biofilm control also relies on well-defined cleaning and sanitizing procedures and the effectiveness of these procedures. Bacteria within biofilms are more difficult to eliminate than free-living cells and once established can act as a source of contamination. Contamination attributed to biofilm development has been reported in general milk processing (e.g. pasteurization and milk transfer line) and the manufacture of cheese, whey and milk powder. Pathogenic micro-organisms, including *Listeria monocytogenes*, *Salmonella typhimurium* and *Yersinia enterocolitica*, will also attach to surfaces in dairy processing environments, e.g. stainless steel. Subsequently, dairy products may be contaminated.

Attachment of micro-organisms may be promoted or inhibited in dairy fluids depending on the composition of the dairy fluid and the type of bacteria. The inhibition of attachment has been reported in the presence of whole milk but enhanced by the presence of lactose and non-casein protein solutions. An association of the bacteria with milk fat globules and the effect of natural antibodies have been suggested as possible reasons for this.

The pasteurizer can be a source of contamination of *Bacillus* spp., especially after non-production days such as at weekends. In addition, the growth of

bacteria, e.g. *Streptococcus thermophilus*, on the surface in the regeneration section of plate heat exchangers can contaminate milk with <100 to >10⁶ bacteria per ml and/or their metabolic products (Figs 4.4 and 4.5). This affects the quality of products manufactured from this milk, but fouling also leads to an increased use of energy, resulting in a decrease in production time, obstruction and corrosion and causing considerable economic loss. The filling machine is a



Fig. 4.4 *Streptococcus thermophilus* bacteria adhered to a plate of a heat exchanger.

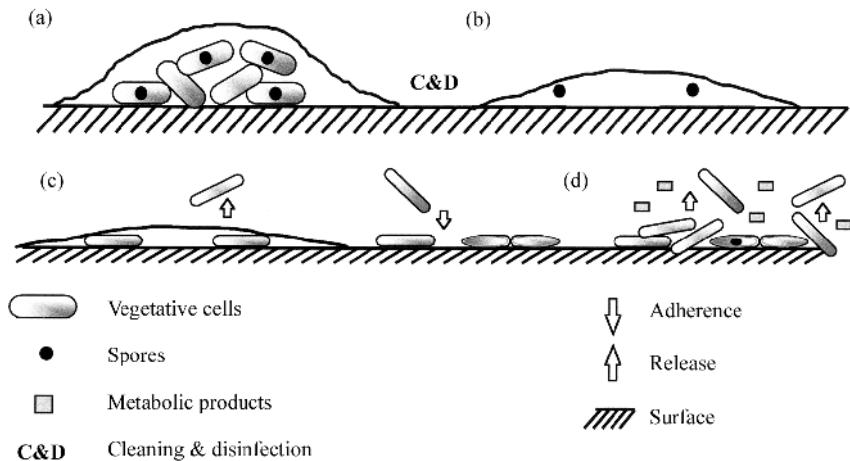


Fig. 4.5 Adherence of bacterial spores on equipment surfaces: (a) vegetative bacteria and spore-forming bacteria adhered to a surface; (b) spores survive cleaning and disinfection; (c) outgrowth of spores and release of vegetative cells into the bulk-phase, i.e. the food product; (d) adherence of bacteria, release of bacteria and metabolic products into the bulk-phase.

significant source of post-pasteurization contamination. The presence of spoilage psychrotrophs (*Acinetobacter*, *Pseudomonas* and *Flavobacterium* spp.) in pasteurized milk is considered to occur after pasteurization and indicates inadequate cleaning.

Packaging material, carton-forming mandrels, filling heads and airborne micro-organisms were identified as major contamination sources. Food-grade paper and board used in the dairy industry are usually of high hygienic quality and microbial counts are well below the limits set by the FDA, $\leq 1 \text{ cfu/cm}^2$ or 250 cfu/g. In a study it was demonstrated that the contamination of the inner surface of cartons intended for liquid foods rarely exceeded 10 cfu per package of one litre capacity. Re-usable milk bottles have been shown to be contaminated by spore-forming organisms such as *B. cereus* in concentrations of <10 to 250 per 100 ml rinsing water.

4.3.3 End-products

Five factors limit the shelf-life of refrigerated pasteurized milk:

- The microbiological quality of the raw milk
- Time and temperature of pasteurization
- Presence and activity of post-pasteurization contaminants
- Types and activity of pasteurization-resistant micro-organisms
- The storage temperature of milk after pasteurization.

The relation between storage temperature and shelf-life of pasteurized milk is well recognized. Low temperatures retard the growth of bacteria and conversely

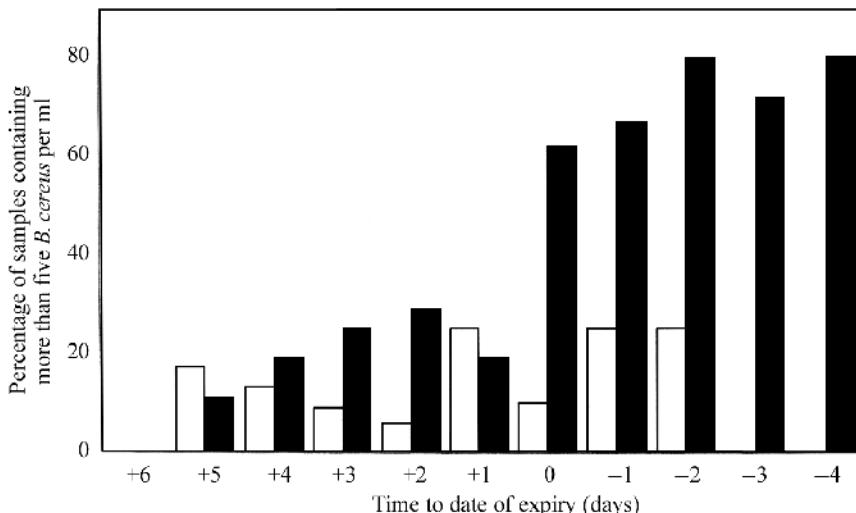


Fig. 4.6 Incidence of *B. cereus* in pasteurized milk (100 ml samples) in households in relation to storage time and temperature of the refrigerator ($\square < 7^\circ\text{C}$; $\blacksquare > 7^\circ\text{C}$).

increase shelf-life. In Fig. 4.6 the percentage of samples containing more than five *B. cereus* per ml in household refrigerators is shown as a function of the temperature measured in the refrigerator and the time to expiry date of the milk. As expected, the level of *B. cereus* present in milk increased with storage time and temperature. The temperature should be maintained at less than 4°C in the distribution chain to reduce growth of psychrotrophs. Monitoring of the temperatures and information on temperature history can be used to identify problem areas and allow improvements to be made.

Training programmes could ensure that all people involved in processing, distribution and handling of milk and dairy products understand the principles of personal hygiene, milk spoilage and the need to keep milk cold constantly. Consumers also have to be educated as to the importance of keeping milk cold. Label information on packages may help to achieve improved quality control of milk and dairy products.

4.4 Future trends

4.4.1 Farm

In order to assure high quality standards for dairy products, it is necessary to manage the whole production chain from farm to consumer. In the whole process, the chains at the beginning of the process, especially the feed, become more and more important.

The automation of raw milk production on farms is increasing. Automatic milking systems have been commercially available since 1992 and sales of such

systems have grown rapidly. In 1999 there were over 300 systems installed; at the end of 2000 there were over 1000 systems in operation on farms in Europe. Benefits of the systems include reduced labour demand, improved animal health and welfare and increased milk yields. However, investigations have shown persistent increased levels of free fatty acids, freezing points, total bacterial counts and somatic cells in milk. With respect to control of the bacteriological quality of the milk, important aspects of automatic milking systems are cleaning of teats and milking equipment and the (direct or indirect) cooling systems. More research is necessary and corrections or improvements should be made to raise the quality of milk.

4.4.2 Product and process development

Consumer demands for healthier foods also influence the development of dairy products. The current generation of dairy products contains those that have been nutritionally improved by enhanced formulations of traditional dairy products. Modifications of dairy products include modifications or reductions in fat, cholesterol, sodium or calories and addition of beneficial components such as calcium.

The dairy industry can meet the needs of consumers and expand the dairy product market by undertaking new approaches to processing and product development. Target areas that have potential are, e.g., traditional products that indulge but balance nutrition, new product concepts utilizing dairy components, food service products, processing and formulation technologies to extend shelf-life to 20–45 days, new packaging strategies, convenience, excellent sensory characteristics and safety. Innovative new technologies (e.g. high-pressure processing, separation technologies) or alternative uses of existing technologies (e.g. steam infusion) can be applied for the development of new products. Biotechnological and separation technologies can provide ingredients, isolated dairy components and bacterial cultures that are important for developing new dairy formulations. Many of the technologies are still capital intensive. Furthermore, safety issues relevant to new formulations and processing conditions for extended shelf-life products will continue to challenge the food and dairy industries.

4.4.3 Monitoring, control and optimization of production processes

The food industry has concentrated on examination of end products for controlling production processes. The accent shifted from analysis at the end of the process to control of the process, by the introduction of Good Manufacturing Practice (GMP) and the Hazard Analysis of Critical Control Points (HACCP) system. In HACCP systems, microbiological methods are needed for, among other things, assessing the quality of the raw materials, detecting micro-organisms in process lines and the environment, and validation and verification. A measuring system (control measure) is necessary to make sure that the critical control points (CCPs) are controlled indeed.

Most ideal is a continuous registration system by means of physical and chemical analyses. Developments in the area of sensors will continue and lead to applications within the food industry within the next 10 years. Classical microbiological tests are unsuitable for quickly obtaining current measuring data and readjusting processes. Therefore, much research has been carried out to improve and develop rapid detection methods for micro-organisms and/or metabolites. Developments in the areas of immunology, molecular biology, automation and computer technology occur at a rapid pace, and can contribute to more rapid, more sensitive and user-friendly methods for the food industry. In a recent study, various microbiological methods were used to monitor the development of micro-organisms during thermization and pasteurization of milk. The results show that some of the methods currently available offer possibilities for application as an 'emergency brake'. The main problem is the sensitivity of the techniques. Levels of 10^4 – 10^5 micro-organisms per ml can be measured, but this is not sufficient to adjust a production process. To permit routine in-process measurements in a production setting, the operation of the equipment will have to be simplified. Monitoring metabolites of micro-organisms is not yet an alternative because the moment anything could be measured, (too) high concentrations of bacteria are already present in the food matrix.

Control and optimization of production processes are of great importance to the food industry as this may lead to improvement of products and processes, and to cost savings. Integrated process and product development by applying rapid detection methods for critical process parameters and predictive models, therefore, is a challenge to the food industry.

In the future (objective) process control systems can be developed by integrating results of (microbiological) analyses and predictive models into process control software. In this way it is possible to adjust processes more efficiently and to respond to deviations more quickly. Computer control, neural networks and fuzzy logic may also be useful to this end.

4.5 Sources of further information and advice

<http://www.nsf.org/>
<http://www.ehedg.org/>
<http://www.3-a.org/>
<http://www.fil-idf.org/>
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