CHAPTER 12

Practical Applications: Prospects and Pitfalls

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INTRODUCTION

Fermentation is one of the oldest technologies used for food preservation. Over the centuries, it has evolved, been refined, and diversified until today, when a large variety of foods are derived from this technology, both in the industrialized and the developing countries, in households, small-scale food industries, and large commercial enterprises. Fermented foods form a major part of the human diet all over the world. In some regions, mainly in African countries, fermentation plays an important role in the nutrition of infants and young children because it is used for the preparation of complementary foods.†

Advances in food science and technology have given rise to a wide range of new food technologies. Nevertheless, fermentation has remained one of the most important food processing techniques throughout human history. Many benefits are attributed to fermentation. It can preserve food (i.e., increase shelf life), improve digestibility, enrich food, and enhance taste and flavor. It is also an affordable technology, and thus is accessible to all populations. Furthermore, fermentation has the potential to enhance food safety by controlling a great number of pathogens in foods. Thus, it makes an important contribution to human nutrition, particularly in developing countries, where economic problems are a major barrier to ensuring food safety.

In general, fermented foods, particularly those produced under controlled conditions, have a good record of safety and are implicated in outbreaks of diseases relatively infrequently. There are, however, more concerns with traditional and artisanal productions, where the application of fermentation is based largely on experience and knowledge gained through trial and error by generations of food producers and households. Such an empirical approach presents a major pitfall. Depending on the process, ingredients, raw material, and environmental conditions, the process may lead to unsafe products.

This chapter reviews the importance of fermentation from a public health and food safety point of view. Specific emphasis is put on problems in developing countries where risks of food contamination and disease are greater, and the potential contribution of fermentation for processing and storage of foods more important. The chapter examines the risks and benefits of fermentation for human nutrition, with a particular focus on the fermentation of complementary

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Note: This chapter is partly based on the report of a Joint Food and Agriculture Organization(FAO)/World Health Organization (WHO) Workshop on fermentation as a household technology to improve food safety held in Pretoria, South Africa, 11–15 December 1995.[18]

†The term *complementary foods* refers to any nutrient-containing foods, be they solids or liquids, other than breast milk, that are given to infants and young children during the period of complementary feeding. This period of complementary feeding corresponds to the period during which other foods are provided along with breast milk.

foods, and looks into the prospects for promoting the technology and improving the safety and nutritional quality of the derived products. It also presents two applications of the Hazard Analysis and Critical Control Point (HACCP) concept to fermented foods as examples.

BENEFITS OF LACTIC ACID FERMENTATION

Depending on the organism used (i.e., molds, yeasts, bacteria), there are different types of fermentation processes. Fermentation mediated by lactic acid bacteria (LAB) is the process that is presently of greatest interest to food safety and public health. Other types of fermentation are also important to public health because they may contribute to food security and overcoming malnutrition. The focus of this chapter is on lactic fermentation.

There is considerable evidence to show that lactic acid fermentation inhibits the growth, survival, and toxin production of a number of pathogenic bacteria[1] (see also Chapter 2). The inhibitory effect of lactic acid fermentation on pathogenic bacteria as well as the spoilage organisms is due to the rapid growth and acid production by LAB, the consequent decrease in pH, and the formation of other antimicrobial factors associated with LAB, such as bacteriocins, hydrogen peroxide, ethanol, and diacetyl.[1,8] The extent to which pathogens are inhibited depends on the organism concerned, the temperature, the amount of acid produced, and the properties of the food (e.g., the buffering capacity). For instance, in cereals and vegetable products that are weakly buffered, an efficient lactic acid fermentation will produce a pH of 4 or less, at which the growth of bacterial pathogens is inhibited and many bacteria die. This aspect of fermentation, as seen in the following paragraphs, is of great importance to public health, particularly in developing countries where safe food storage by cold or hot holding is difficult because of socioeconomic constraints.

Lactic acid fermentation has also been associated with the reduction of certain naturally occurring toxins in plant foods, notably a reduction of cyanogenic glycosides that are present in cassava. Cassava is a major staple in Africa and, due to the presence of these compounds, it has been associated with several health problems.[51] Although the degradation of the cyanogenic glycosides is mediated by endogenous plant enzymes, microbial activities carried out during fermentation contribute to the detoxification process by softening the plant tissues (see Chapter 4).

Nutritional benefits have also been attributed to fermentation. Some fermentation processes lead to enhanced digestibility of carbohydrates as a result of degradation of oligosaccharides and dietary fiber, particularly prevalent in foods of plant origin. The use of amylase-rich flour (ARF) combined with a small amount of a lactic acid starter culture can increase the nutrient density of starchy foods while keeping a semi-liquid consistency. This makes the fermented food particularly interesting as complementary food for infants and young children. Fermentation also provides an optimum pH for the activity of phytase and the degradation of phytate. The degradation of phytate increases the amount of soluble iron several fold, thereby increasing its bioavailability. Lactic fermentation also decreases the tannin content of cereals and, in this way, has a positive impact on mineral absorption and the protein digestibility of cereals.

Over and above the influence on pathogenic bacteria and toxic compounds leading to improved food safety and quality, specific beneficial health effects such as prophylaxis against *Escherichia coli* and other pathogens and hypocholesterolemic and anticarcinogenic effects have also been attributed to some selected strains of LAB. A number of such organisms with claimed health-related properties are already used in food production and are available in many countries. The use of probiotics raises two fundamental questions: (1) Are the organisms used safe? and (2) Are the health claims valid? The former has been discussed in Chapter 11 of this book. The latter is the subject of extensive research. A number of studies have shown some specific health benefits with regard to lactose intolerance and gastrointestinal disorders. So far, there is no conclusive explanation for the underlying molecular mechanisms. Also, there

are a number of claims that have not been substantiated and need further investigation.[13,22]

Advances in genetic modification open new doors to fermentation technology.[34] Genetic modification may be carried out on the substrate (e.g., plants) or on the microorganisms used for fermentation. Fermentation may also be used for the production of enzymes with enhanced properties and improved quality (see Chapter 10). These can have significant benefits in terms of improving the fermentation process and the quality and safety of the final product.

Genetic modification can be used in different ways to improve the fermentation process and to enhance the quality and safety of the final product. For instance, starter cultures may be improved with regard to their rate of growth, potential for lactic acid production, salt tolerance, inability to metabolize organic acids, resistance to bacteriophage, production of flavor compounds, and so on.[6,8] Genetic modification may also be used to remove or reduce hazards in the raw material, as in the production of a variety of cassava containing less cyanogenic glycosides.

As discussed in Chapter 10, the prospect of using genetic modifications for better control of fermentation processes and to enhance the safety and quality of derived products has two preconditions: (1) an adequate method for assessing the safety and nutritional implications of these products and (2) consumer acceptance. FAO, WHO, and other organizations have developed strategies to assess the safety of such foods,[17,50] and further work in this area is planned. To gain consumer acceptance, genetic modification needs to be considered in the broader perspectives of risks and benefits for the consumer, environment, socioeconomic implications (in both developing and industrialized countries), and, in general, the need for such technology. The prospect of using genetic modification technology for improving fermentation processes relies on an appropriate risk communication with consumers.

It should be mentioned that genetic modification can also occur spontaneously in nature and lead to new starter strains. Conventional methods of screening and selection of new starter strains with desirable properties have been used for the production of products with enhanced quality.[34]

PITFALLS OF FERMENTATION

As simple as it may seem, fermentation is also a sensitive and complex technology. The safety and quality of the final product depend on a number of factors, which need to be carefully controlled. These factors are

- quality and safety of raw materials, including the initial level of contamination
- levels of environmental hygiene and sanitation
- quality and safety of starter culture
- safety of metabolites
- processing conditions and degree of acidity achieved

In certain applications, particularly in the context of cottage industry or household application of fermentation, it may be difficult to control these factors adequately. Not infrequently, fermented products have been incriminated in infections or intoxications. In certain settings, inadequate fermentation has been a major cause of food-borne intoxications. For instance, in China, 2,861 cases of botulism (745 outbreaks) were reported in the year 1989, causing some 421 deaths. The major implicated foods were homemade fermented bean products. Fermented fish and salmon eggs are a significant cause of botulism occurring among the native Alaskan Inuit population in the North American continent. Fermented Inuit products such as white whale meat, seal flippers, or salmon eggs lack fermentable carbohydrates and therefore do not undergo a sufficient pH reduction to prevent toxigenesis.[20] Cheeses of different categories have also been implicated in outbreaks of various types of food-borne diseases.[24] Examples of other documented outbreaks of illness are listed in Table 12–1. It is likely that outbreaks of food-borne disease caused by fermented foods produced under unhygienic conditions or with faulty handling of the products occur more frequently than are officially reported. However,

Table 12–1 Examples of Food-Borne Disease Outbreaks Associated with Fermented Foods

Implicated Food	*Causative Agent*	*Number of Cases*	*References*
Vegetables			
Paste of soybeans and wax gourds	*Clostridium butyricum*	6	32
Sauerkraut	Histamine		30
Milk products			
Curd (yogurt)	*Campylobacter jejuni*	160	7
Yogurt	*Clostridium perfringens*	167	33
Hazelnut yogurt (hazelnut puree was contaminated)	*Clostridium botulinum*	27	36
Sour milk	*Clostridium botulinum*	11	41
Yogurt	*Escherichia coli* O157	16	35
Meat products			
Semi-dry sausages	*Escherichia coli* O111:NM	23	10
Pork (salami)	*Salmonella anatum*	52	4
Pork (labh, raw and nahm, fermented)	*Trichinella*	27	26
Salami stick	*Salmonella typhimurium*	85 (including 13 secondary cases)	12
Salami	*Escherichia coli* O157	23	11
Sausages (Lebanon bologna)	*Salmonella typhimurium*	26	38
Fish			
Fish (seal flipper)	*Clostridium botulinum*	1	39
Fish (beaver tails)	*Clostridium botulinum*	7	40
Fish (salmon fish heads)	*Clostridium botulinum*	8	39
Salmon eggs	*Clostridium botulinum*	15*	20
Cheeses			
Soft cheese (Mexican-style)	*Listeria monocytogens*	142	24, 27
Soft cheese	*Salmonella berta*	82 (including 3 secondary cases	15
Cheese	*Salmonella enteritidis*	≅700	9
Soft cheese	*Salmonella dublin*	42	28
Goat milk cheese	*Salmonella paratyphi*	273	14
Cheddar cheese	*Salmonella heidelberg*	339† (28000–36000 est.)	19
Mozzarella cheese	*Salmonella typhimurium*	321	3
Cheese	*Escherichia coli* O157	22	44
Cheese (Brie, Camembert, Coulommiers)	*Escherichia coli* O124:B17	387 est.	29
Cheese (Brie, Camembert)	*Escherichia coli* O27:H20	170	3
Cheese (Brie, Camembert)	*Clostridium botulinum*	27	37
White processed cheese	*Streptococcal pharyngitis*	197	5
Mexican-style soft cheese	*Streptococcus zooepidemicus*	16	16
Mexican-style soft cheese	*Brucella melitensis*	31	3
Hand-pressed direct set cheese	*Staphylococcus aureus*	16	3
Cheese	*Staphylococcus aureus* and *Shigella sonnei*	≥ 50**	40
Swiss cheese	Histamine	6	43

*15 cases involved in 7 outbreaks between 1971–1984.

†339 cases where reported in Colorado. From the attack rates noted (28–36%) and the amount of cheese presumably consumed (2830 kg), it is estimated that between 28,000 and 36,000 persons were affected in total.

**Bacillary dysentery, which affected numerous persons who had eaten various French cheeses purchased at a Paris airport in 1982, was reported by several Scandinavian countries.

weaknesses in the food-borne disease surveillance system, particularly in regard to household food preparation, do not provide for identification and reporting of these outbreaks.

Although LAB can inhibit the growth of certain food-borne pathogens, care should be taken to ensure that the raw material is of high hygienic quality and the risk of contamination from the environment is minimized. A number of food-borne hazards are not controlled by lactic acid fermentation and so present a serious threat to health if they persist in food. For instance, enterohemorrhagic *E. coli* has shown patterns of acid resistance and may survive certain fermentation processes. Yogurt and fermented meat have been recognized as potential vehicles of enterohemorrhagic *E. coli* infection. Food and waterborne viruses, a relatively frequent cause of gastroenteritis, may survive high levels of acidity. For instance, Simian rotavirus has been shown to survive a high level of acidity during 24 hours of storage in model fermented foods[53] (see Chapter 8). There is little information on the effect of fermentation on parasites, such as *Cryptosporidium, Giardia lamblia,* and food-borne trematodes. The cysts or metacercariae of these organisms often show resistance to adverse conditions (see Chapter 9). Thus, the possibility that they may survive fermentation should not be excluded. Most toxins produced by algae, bacteria, and molds are also unaffected by fermentation (see Chapter 5).

In view of this, it is important not to rely only on the fermentation step *per se* to eliminate or reduce hazards to safe levels. To ensure safety, it is important to design the process in such a way as to (1) prevent contamination, (2) eliminate or reduce the hazards as much as possible, and (3) control the growth of pathogens. For this reason, fermentation steps frequently need to be combined with other process operations such as soaking, cooking, and so forth. Lactic acid fermentation also has a limited effect on antinutritional factors, such as protease inhibitors and lectins, and other process operations such as heat treatment should be used to destroy these antinutritional factors.

There are also some concerns with the organic acids that are produced during lactic acid fermentation. Microbially produced lactic acid is usually a mixture of the optical isomers L(+) and D(-) lactic acid. D(-) lactic acid cannot be metabolized by humans. Excessive production and intake of D(-) lactic acid may result in acidosis, a disturbance of the acid-alkali balance in the blood. However, very little is known about the "toxicity" of D(-) lactic acid for malnourished or sick children, and little data are available on the D(-) lactic acid content of fermented foods prepared at the household level. Further studies in this area are needed to elucidate the role of D(-) lactic acid and methods to control its production.[1,2,45,54]

Histamine poisoning is also a potential problem with fermented foods. Although histamine poisoning has been commonly associated with the consumption of scrombroid-type fish such as tuna and mackerel, certain fermented foods such as wine, soy sauce, and, in particular, cheese, may also present a risk. Poisoning due to histamine or other biogenic amines is caused by an accumulation of histamine or other type of biogenic amines (e.g., tyramine, phenylethylamine) following metabolic activity of decarboxylase-positive LAB such as *Lactobacillus büchneri.* However, the problem can be controlled to a large extent by observing good manufacturing practice during production. For instance, during the manufacture of cheese, the control of factors such as hygienic quality of milk and temperature of storage minimize the risk of histamine formation[42] (see Chapter 6).

IMPORTANCE OF FOOD FERMENTATION IN PUBLIC HEALTH

Food-borne diseases are a major public health problem all over the world, in both industrialized and developing countries. The industrialized countries, benefiting from a higher standard of living, good water supply and sanitation, and technologies for processing and preserving foods, have succeeded in combating many food-borne infections. This has resulted in a reduction of a great number of food-borne infections such as typhoid fever, cholera, and shigellosis. Nevertheless, food-borne diseases remain a widespread public health problem, and statistics in these countries indicate that possibly up to 30%

of the population may suffer from a food-borne illness annually.[31]

It is the developing world that bears the brunt of the problem. Although statistics on the incidence of food-borne diseases are not available, the high prevalence of diarrheal diseases in these parts of the world, particularly in infants and young children, is an indication of an underlying food safety problem. In 1997, 4,000 million cases of diarrhea were estimated to occur in the world.[52] Approximately 1,500 million episodes of diarrhea occur annually in children under the age of five, and more than 1.8 million children die as a result. Indirectly, diarrheal diseases kill many more children because they are one of the major underlying factors of malnutrition. It is estimated that annually, some 13 million children under the age of five die from the associated effect of malnutrition.

The etiological agents responsible for food-borne diseases are broad and include bacteria, viruses, and parasites, all of which have been discussed here individually in their respective chapters.

The sources of contamination of food are diverse and include polluted water, night soil, dust, flies, domestic animals, dirty utensils, and food handlers. Raw foods themselves may also be a source of contaminants because many foods harbor pathogens or come from infected animals. Moreover, during the preparation process, there is an added risk of cross-contamination. However, one of the major factors leading to food contamination is time-temperature abuse during food preparation and storage with the result that pathogens survive, grow, and produce toxins (Figure 12–1).

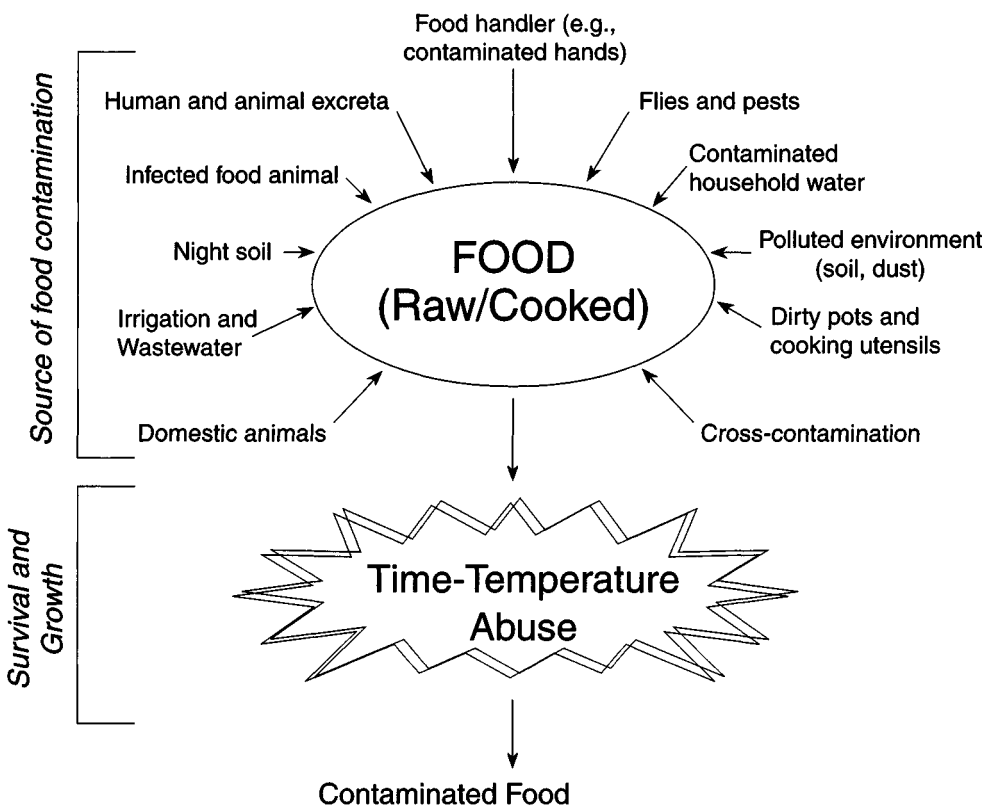


Figure 12–1 Sources of food contamination

In addition to agents of diarrheal diseases, food may also be a vehicle for chemical hazards, whether naturally present or contaminating the food as a result of poor agricultural practices or environmental pollution as dealt with in Chapters 4, 5, and 6. Depending on the dose, chemical hazards may lead to acute intoxication or long-term health problems such as cancers and other chronic diseases. Food may also contain antinutritional factors such as enzyme inhibitors, phytates, lectins, and polyphenols, which interfere with the digestion, absorption, or other aspects of metabolism of nutrients in foods.

Food processing technologies are applied for a variety of reasons. They may be applied to render food more digestible or edible, to retain or enhance sensory quality, to increase shelf life, to improve nutritional quality, and/or to render food safe. Food fermentation has proved to possess many of these features; in this way, it is an important food technology. However, its role in improving the nutritional quality of foods, particularly complementary foods, and preserving foods and preventing growth of most pathogenic organisms make this technology particularly important from a public health point of view.

Fermentation is particularly important for food safety and for the prevention of diarrheal diseases in the developing regions. Although the application of basic rules of food hygiene can prevent a great proportion of diarrheal diseases, in the developing countries, the application of these rules is sometimes hampered by socioeconomic constraints such as inadequate supply of safe water, lack of knowledge or facilities for safe preparation and storage of food (e.g., refrigeration, fuel for hot holding or thorough reheating), and lack of time to prepare food properly before each meal. As a result, some households, particularly low-income ones, are simply not able to apply essential food safety principles, such as feeding infants with freshly prepared foods, chill storage, hot storage, reheating of stored foods, and so forth.

Thus, fermentation provides an economic means of preserving food and inhibiting the growth of pathogenic bacteria even under conditions where refrigeration or other means of safe storage are not available. At the same time, it can enhance the nutritional quality of some foods. In several African countries, the technology is used in particular in the preparation of foods for infants and young children.[2] For instance, in Kenya, Nigeria, the United Republic of Tanzania and Uganda, it is customary to give infants fermented cereals, or root-crop products. Fermentation is also used to produce beverages. Again, in areas where the safety of the water cannot be ensured, fermentation processes contribute to reducing the risk of waterborne diseases.

PRACTICAL INTERVENTION TO ENHANCE SAFETY OF FERMENTED FOODS

To take advantage of the benefits that fermentation offers and, at the same time, to minimize its risks, it is important to examine the fermentation process carefully and to develop a plan where hazards associated with the different production steps are considered and controlled. In Chapter 3, the concept of Hazard Analysis and Critical Control Point (HACCP) as a method of food safety assurance was explained, and a plan for a fermented indigenous food that is common in Africa and prepared at household level was presented. Other examples of the application of the HACCP concept to fermented African foods have also been published elsewhere.[46–48] In this section, a second example of an indigenous African food, *togwa,* is presented to illustrate how the concept of HACCP can be used to evaluate a food production or preparation process in order to identify possible safety problems and how they may be controlled.

This study illustrates that the process of togwa preparation, as described here, leads to a high-risk product. The risk lies in the fact that "power flour" (an ARF obtained from germinated seeds), which may be contaminated, is added after cooking. If fermentation fails, proliferation of contaminant bacteria may occur and acid-resistant pathogens may survive. Possible options to reduce risks include accelerated fermentation by back-slopping or use of starter culture.[21] Extreme care should be taken to prevent

postcooking contamination. The HACCP study of this product identifies numerous critical control points (CCPs) reflecting the high degree of concern. The reasons for concern are that (1) the food is intended primarily for infants and young children, who, due to their susceptibility to infections, require great care in the preparation of their food; (2) a wide range of hazards have been considered in this study; and (3) the preparation is envisaged in a household setting with rudimentary environmental conditions. Because the risk of contamination in this setting is higher, several steps in the preparation likely to prevent or control contamination need to be carefully controlled.

A second example shows an HACCP study for the production of a traditional cheddar cheese. Cheddar cheese has been implicated in outbreaks of salmonellosis and *Staphylococcus aureus* intoxication.[19,25,49] In one outbreak, the problem was due to improper application of the HACCP system (i.e., inadequate pasteurization and a lack of corrective action when the monitoring results indicated failure in reaching critical limits).[49]

Both examples also underline the importance of good hygienic practice (GHP) as a prerequisite for the production of fermented foods, particularly with regard to the prevention of contamination. In the case of cheddar cheese, it can be seen that after the pasteurization step, there is no other step that can eliminate pathogens. Therefore, GHP is extremely important to prevent recontamination.

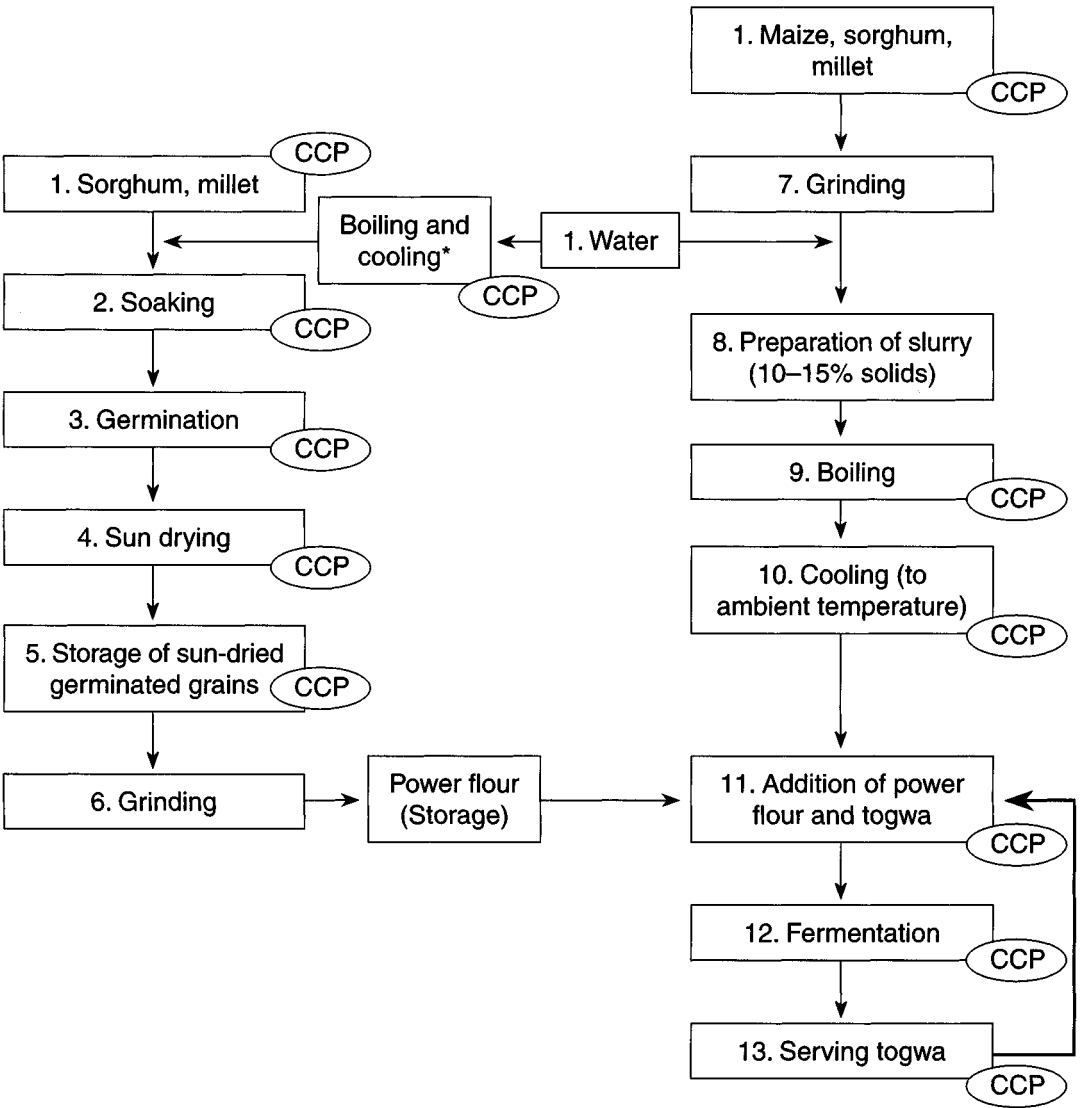
HACCP Study of Togwa*†

1. **Product description**—Togwa is prepared by many tribes in Tanzania, and there are variations in its production. The preparation involves mixing cooked cereals (e.g., maize, sorghum) with germinated cereals (e.g., finger or bulrush millet or sorghum). Fermentation takes approximately 9–12 hours, depending on whether or not an old batch is used as a starter culture. If an old batch or power flour is used, the fermentation is completed in approximately 6–9 hours.
2. **Intended use**—Most fermented gruels in Tanzania remain edible for one to two days. Beyond this period, the product is too sour, or gives off an unpleasant odor. Fermented gruels are traditionally given to children less than five years of age.
3. **Flow diagram**—Figure 12–2 shows the flow diagram of togwa.
4. **Hazards of concern**—Hazards considered in this context include biological (e.g., bacteria, viruses, parasites), chemical (e.g., contaminants, mycotoxins), and physical agents.
5. **Identification of hazards, control measures, and CCPs**—Table 12–2 shows hazards associated with each step in the preparation of togwa and of power flour, which is an essential ingredient in this gruel.
   a. *Raw material:* Major hazards in maize, millet, and sorghum are toxins (e.g., aflatoxin produced by molds, and agrochemicals). For prevention of the activity of toxigenic molds during storage, the raw material should, as far as possible, be stored under appropriate conditions. When the ambient temperature and humidity are high, storage time should be limited. Insofar as agrochemicals are concerned, households cannot do much except to get assurance from suppliers concerning the safety of the products. The possibility of accidental contamination of grains during storage with agrochemicals should be prevented.

      The grains may also contain foreign matter such as stones and insect fragments. These will not be eliminated at a later step, so it is important that households thoroughly clean the raw material.

*Application of the HACCP system has been simplified and adapted to household conditions. Although the same approach can be used for production on a cottage and industrial scale, the requirements in terms of critical control points, critical limits, and monitoring procedures may be different and more severe.

†Model HACCP plans are not appropriate for use until validated for a specific food and food process.



Note: Numbers correspond with those in Table 12–2.
*Although this step was not part of the original togwa preparation, it was added after the HACCP study because boiling water was identified as being essential and needed to ensure the hygienic quality of power flour.

Figure 12–2 Flow diagram of togwa. The high number of CCPs identified in this study is due to the fact that the study is considered in poor hygienic conditions.

Water used in the preparation of slurry may be contaminated. Boiling the slurry (see step 9) will eliminate eventual pathogens. Therefore, water used for the preparation of slurry is not a CCP, although as part of a good hygienic practice, safe water should be used as far as possible in the preparation of togwa and for washing hands and utensils. On the other hand, the use of safe water in the preparation of power flour is essential because this

Table 12–2 HACCP Study of the Preparation of Togwa in Households

Step	*Hazards*	*Control Measures*	*CCPs*	*Critical Limit**	*Monitoring Procedure*	*Corrective Actions*
1. Raw material i) Maize Sorghum Millet	a. Mycotoxins	a. i) Obtain assurance from supplier of adequate preharvest and postharvest handling of grains	a. Yes	a. i) No moldiness, good smell	a. Observation, smelling	a. Discard the raw material and change supplier
		ii) Store grains in dry (and if possible cool) area, limit storage time		ii) Short storage time, adequate temperature humidity of storage area	Time keeping (measurement of temperature or humidity if possible)	Utilize the raw material as quickly as possible.
	b. Agrochemicals	b. Obtain assurance from supplier of adequate preharvest and postharvest handling of grains	b. No			
	c. Pathogens: *Bacillus cereus, Salmonella, Escherichia coli*	c. Heat treatment, fermentation	c. No			
	d. Physical: insects and stones	d. Manual cleaning	d. Yes	d. No visible stones	d. Observation	d. Reclean.
1. Raw material ii) Water	a. Chemical contaminants, depending on the source	a. Obtain assurance about the source of water; use only safe water	a. Yes	a. Clear, free of odor and off taste	a. Observation, smelling, and tasting	a. Use another source of water.
	b. Pathogens (e.g., *Escherichia coli, Campylobacter, V. cholerae, Salmonella, Cryptosporidium, Giardia lamblia, Entamoeba histolytica*) Rotavirus	b. If safe water (i.e., filtered and disinfected) is not available, boil the water	b. Yes for step 2; No for step 8	b. Bubbles	b. Observation	b. Reboil.

*In this study, the preparation of togwa is considered in a household environment; therefore, the critical limits are expressed in qualitative values.

continues

Table 12–2 continued

Step	*Hazards*	*Control Measures*	*CCPs*	*Critical Limit**	*Monitoring Procedure*	*Corrective Actions*
2. Soaking	Growth of microorganisms	As far as possible at low temperatures	Yes	Water should remain free from odor or foam	Observation, smelling	Refresh water.
3. Germination	Growth of microorganisms (e.g., toxigenic molds)	As far as possible at low temperature	Yes	Grains should remain free of moldiness	Observation	Remove moldy grains.
4. Sun drying	a. Contamination and introduction of foreign matter	a. Protect the sprouts	Yes	a. No foreign matter	a. Observation	a. Clean if possible. If not, discard
	b. Inadequate drying may lead to growth of microorganisms during storage	b. Ensure thorough and fast drying		b. Sufficient time, adequate exposure to sun, dry ambient conditions, adequate air circulation, no mold	b. Time keeping, observation	b. As long as there is no mold growth, redry under proper conditions; otherwise, discard.
5. Storage of sun-dried germinated grains	a. Contamination/ introduction of foreign matter	a. Protect the grains	Yes	a. No foreign matter	Observation	a. Clean if possible. If not, discard
	b. Growth of toxigenic molds, if the moisture content is high	b. Keep dry		b. Dry conditions of storage, no mold		b. Discard
6. Grinding to power flour	Introduction of filth, dirt and foreign matter	Use clean and properly maintained equipment	No			
7. Grinding	same as step 6					

continues

Table 12–2 continued

Step	Hazards	Control Measures	CCPs	Critical Limit*	Monitoring Procedure	Corrective Actions
8. Slurry preparation	Contamination with pathogens through utensils and/or water	Use clean utensils and safe water	No			
9. Boiling	Survival of pathogens	Thorough boiling	Yes	Bubbles	Observation	Reboil.
10. Cooling	a. Growth of bacterial spores	a. Ensure rapid cooling	Yes	a. Short time, room temperature within four hours	a. Time keeping	a. Reboil.
	b. Contamination	b. Protect the porridge during the cooling process		b. No foreign matter	b. Observation	b. Depending on the nature of contamination, either clean, reboil or discard
11. Addition of:						
a. power flour	a. Contamination with pathogens by power flour	a. Ensure hygienic quality of power flour	Yes	a. Power flour of high hygienic quality	a. & b. Observation	a. & b. Use another power flour or togwa.
b. Togwa	b. Contamination with acid-resistant pathogens by togwa	b. Ensure the safety of previously prepared togwa		b. Absence of disease upon consumption of previously prepared togwa		
12. Fermentation	a. Growth and formation of toxin by *Staphylococcus aureus*	a. Rapid fermentation	a. Yes	a. Acid taste and characteristic odor within 24 hours	a. Observation	a. Discard the material.
	b. Survival of acid-tolerant pathogens	b. Minimize contamination with acid-tolerant pathogens (see step 11)*	b. No			
13. Serving	Recontamination with pathogens by hands, utensils, and environment	Wash hands and use clean utensils	Yes	Hands properly washed with soap and clean water	Observation	Reheat the food thoroughly.

*As there is no critical control point in the subsequent steps that would ensure the killing of acid-tolerant pathogens surviving the fermentation step, the present process of togwa preparation may lead to a high-risk product.

process does not include a step that would ensure the elimination of pathogens introduced through the raw materials (i.e., water, sorghum, or millet). In addition, pathogens introduced through the raw material may proliferate during the soaking and germination periods and may also survive the sun-drying stage. Therefore, the safety of water used in power flour preparation is critical for minimizing contamination. The hygienic quality of the power flour is particularly important for the safety of the final product because there is no step that would ensure the killing of acid-resistant pathogens after the power flour has been added. Therefore, after the HACCP study, it was suggested to add a boiling step in the preparation of power flour to ensure safety of water and minimize the contamination of power flour.

b. *Soaking:* During the soaking period, bacterial growth will occur. The use of safe water may minimize the final bacterial load. If at all possible, soaking should take place at low temperatures in order to minimize the growth of microorganisms.
c. *Germination:* This usually takes place in layers a few centimeters thick, spread on mats or leaves, and covered with leaves, mats, or gunny sacks to avoid excessive dehydration. From time to time, the material must be aired and mixed while checking and adjusting the degree of grain humidity. Further contamination by mats and microbial growth can occur at this stage. Microbial growth, particularly with respect to toxigenic molds, can also occur. As far as possible, germination should be carried out in cool conditions in order to minimize microbial growth.
d. *Sun drying:* During this step, the cover is removed and the sprouted grains are spread on mats or bamboo trays to dry in the sun. This may take a few days. All kind of contaminants and foreign matter can fall into the sprouted grains if they are not well protected. In addition, insufficient drying may lead to a microbially unstable product during subsequent storage. Thorough drying is critical for the stability of the product.
e. *Storage of sun-dried germinated grains:* Hazards associated with this step are contamination (e.g., by rodents and other animals during storage) and microbial growth, particularly toxigenic molds, if the germinated grains are not properly dried or are kept in humid conditions.
f. *Grinding to power flour:* This step may introduce dirt and foreign matter into the product. As part of a good hygienic practice, this should be avoided as far as possible by using clean and properly maintained equipment. However, it is unlikely that this step will introduce any major health hazard.
g. *Grinding:* Same as step f.
h. *Preparation of slurry:* Except for the safety of water and cleanliness of utensils, no other major hazard is associated with this step. Because the subsequent boiling step will kill the pathogens that may have been introduced at this step, it is not considered a CCP. Nevertheless, as part of a GHP, households should use clean utensils and safe water as much as possible.
i. *Boiling:* Boiling should be thorough in order to gelatinize all the starch. This step is also essential to kill nearly all pathogens (bacterial spores may survive).
j. *Cooling:* The pot should be covered to protect against dirt or other foreign matter falling in. It is important that cooling is carried out as fast as possible. Prolonged cooling may present an opportunity for bacterial spores to grow. When large quantities of togwa are prepared, the cooling time can be

reduced by dividing the togwa into small portions.

k. *Addition of power flour and togwa:* Two ingredients may be added here to initiate the fermentation, that is, power flour and previously fermented togwa. The contamination brought about by power flour is diverse and difficult to control. Viruses and other acid-tolerant agents are of particular concern here. The togwa starter culture is quite acidic, and thus will contain only those contaminants that are acid tolerant.

During the fermentation that follows, acid-sensitive pathogens may be killed. However, acid-tolerant pathogens may survive. A power flour prepared under hygienic conditions may minimize the contamination of togwa. However, in the absence of a final killing step such as thorough reheating, the presence of acid-tolerant pathogens in the final product may not be excluded.

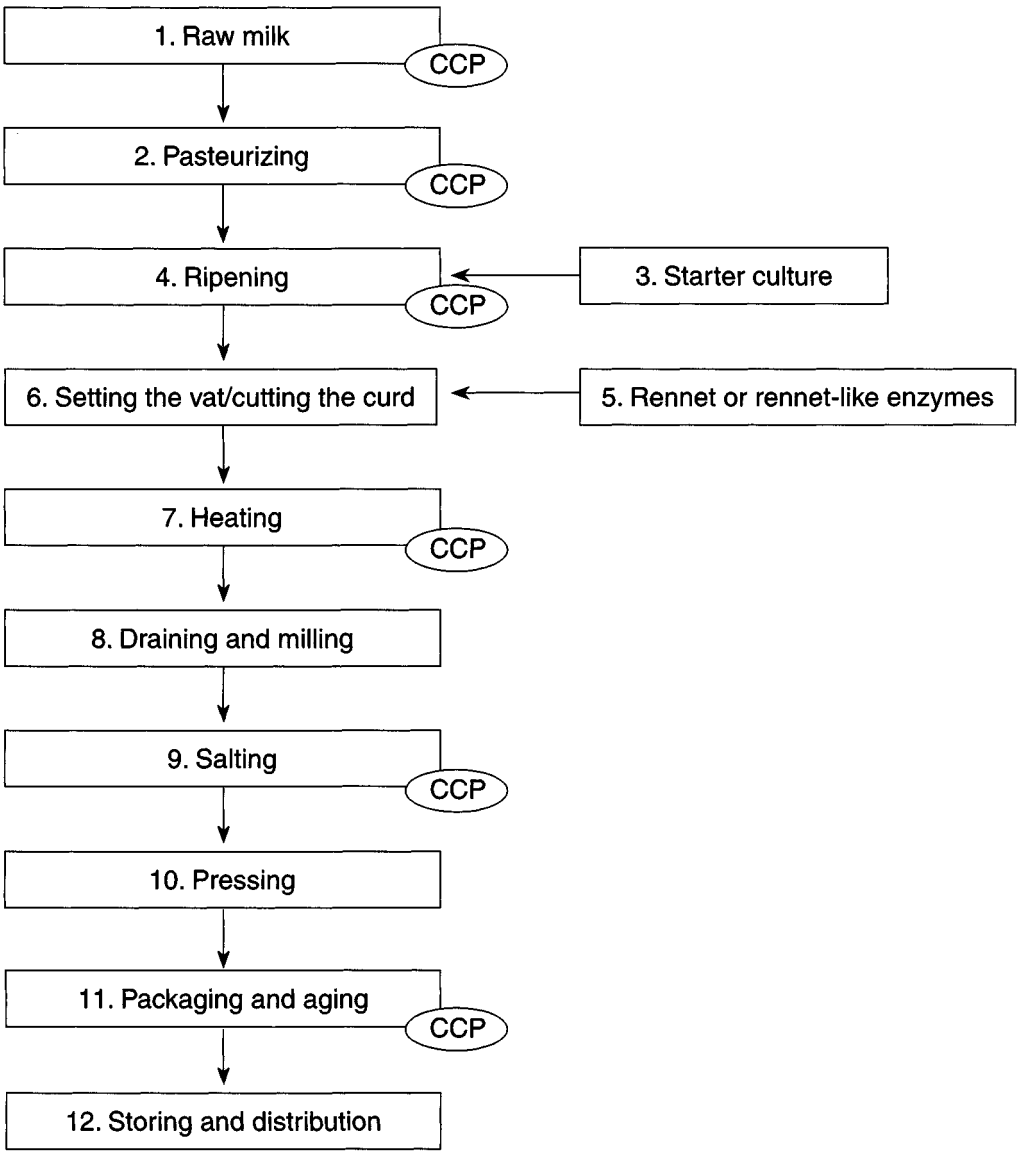
l. *Fermentation:* During fermentation, a rapid dominance of LAB may be expected. This is supported by the short fermentation time to reach the required acidity. A rapid fermentation is critical for killing acid-sensitive pathogens and for preventing bacterial growth and the production of toxin. The addition of togwa enhances fermentation and may be beneficial provided it does not introduce acid-resistant pathogens.

m. *Serving:* It is important to ensure that pathogens are not re-introduced into togwa by dirty hands and utensils. Therefore, these have to be washed carefully with safe water. Depending on the hygienic measures taken to prepare togwa, the final product could be more or less contaminated because there is no final CCP that would ensure the killing of acid-resistant pathogens. Thorough reheating would greatly contribute to the safety of the final product. However, implications in terms of textural and other changes should be considered because the final product may become unacceptable to the consumer.

HACCP Study of Traditional Cheddar Cheese*[23]

1. **Product description**—Cheddar cheese is a hard-pressed cheese that is originally from the United Kingdom. It has a firm body and closed texture. It is composed of approximately 37% water, 33% fat, 25% protein,1% lactose, and 4% ash. The curd is textured after cutting and scalding. It is then milled and salted and is pressed in a mold. The cheese is wrapped in firm blocks and stored at approximately 4–6 °C. The cheese is stored from a few weeks to several months, during which time it ripens and develops its flavor (mild cheese up to 3 months; medium cheese 6 months, and mature cheese 8–12 months). Starter culture used for the production of cheddar cheese consists of *Lactococcus lactis* subsp. *lactis* and *Lc. lactis* subsp. *cremoris.* Other organisms that also play a role during ripening are *Lactobacillus casei*, *Lb. plantarum,* and *Lb. brevis.*
2. **Intended use**—General population, including vulnerable groups.
3. **Flow diagram**—Figure 12–3 shows the flow diagram of traditional cheddar cheese manufacture.
4. **Hazards of concern**—Major hazards are of bacterial origin. Several pathogens such as *Mycobacterium tuberculosis, Brucella abortus, Salmonella, Campylobacter,* and *Listeria monocytogenes* may survive in cheddar cheese. Other hazards of concern are antibiotic residues and histamine.
5. **Identification of hazards, control measures, and CCPs**—Table 12–3 shows

*Model HACCP plans are not appropriate for use until validated for a specific food and food process.



Note: Numbers correspond with those in Table 12–3.

Figure 12–3 Flow diagram of traditional cheddar cheese

hazards associated with each step of the production of cheddar cheese.

- *Raw material:* Raw milk when received by a dairy may be contaminated with a wide range of pathogens, as mentioned above, as well as spoilage organisms. In addition, milk may contain antibiotic residues. Hygienic handling of milk at the farm and during transport is essential to prevent con-

Table 12–3 HACCP Study of the Preparation of Traditional Cheddar Cheese in Industrial Setting

Step	*Hazards*	*Control Measures*	*CCPs*	*Critical Limit*	*Monitoring Procedure*	*Corrective Actions*
1. **Raw material: milk**	a. Pathogenic bacteria: *Mycobacterium* spp., *Brucella* spp, *Campylobacter, Listeria, Salmonella*	Hygienic collection at the farm and transport Refrigeration (maximum temperature 7 °C)	a. No			
	b. Antibiotic residues	b. Control of incoming milk	b. Yes	b. According to Codex Alimentarius Commission	b. Chemical or microbiological tests	b. Reject the milk.
2. **Pasteurization**	Survival of pathogens	Heating	Yes	72 °C for 15 seconds or equivalent	Temperature, time	Repasteurize.
3. **Starter culture**	Presence of hazards in the starter culture	Assurance by the supplier of the starter culture on the quality	No			
4. **Ripening** (addition of the starter culture)	a. Contamination with pathogens	a. Cleaning and disinfection of vat and other equipment	a. No			
	b. Insufficient fermentation	b. Follow instructions of manufacturer of starter culture; use appropriate proportion of starter culture and milk; keep milk warm	b. Yes	b. 30–31 °C, development of acidity of 0.14% before step 8. Proportion of starter culture and milk according to the instructions of the manufacturer	b. Temperature titratable acidity	b. Correct the temperature and the amount of starter culture.
5. **Rennet or rennet like-enzymes**	Presence of pathogens	Assurance of quality by the supplier	No			

continues

Table 12–3 continued

Step	*Hazards*	*Control Measures*	*CCPs*	*Critical Limit*	*Monitoring Procedure*	*Corrective Actions*
6. Setting the vat and cutting the curd	Contamination with pathogens, of utensils used for adding the rennet	Cleaning and disinfecting of cutting knives and hygienic practice	No			
7. Heating	Arresting the fermentation	Control temperature of heating	Yes	37 °C	Temperature	Readjust the temperature.
8. Cheddaring (draining and milling)	Contamination of utensils and environment with hazards	Hygienic practice	No			
9. Salting	Growth of *S.aureus* at subsequent steps	Adjusting the salt level and its distribution	Yes	1.7 %	Amount of salt added	Readjust the proportion of salt and curd.
10. Pressing	Contamination of the press with pathogens	Strict hygiene of the equipment and personnel	No			
11. Aging	Growth of *Staphylococcus aureus;* growth of decarboxylase positive lactic acid bacteria and formation of histamine	Adequate storage (see also step 8)	Yes	5 °C	Temperature	Correct the temperature of storage. Consider removal of the product if the loss of temperature control has exceeded several hours.
12. Storing and distributing	Mold growth	Adequate packaging, refrigeration, and rapid distribution	No			

Note: This table was adapted from ICMSF.[23]

tamination and keep the level of pathogens possibly present to a minimum. To ensure that raw material is of adequate hygienic quality, the milk may be tested for its bacterial load with methylene blue or by some other rapid test. The receipt of the raw milk is a CCP for antibiotic residues because these will not be eliminated by further processing of the milk into cheese.

- *Pasteurization:* The pasteurization step is a CCP because it is essential for the elimination of pathogens that may be present in the raw milk. It will also reduce the number of decarboxylase-positive LAB able to produce histamine.
- *Starter culture:* The contamination of starter culture has in the past caused food-borne disease outbreaks. Therefore, the possibility that the starter culture may be contaminated or be of poor quality (leading to poor fermentation) should be considered, and assurance should be obtained from the supplier of its quality.
- *Ripening of the milk:* At this stage, the milk is cooled to 30–31 °C and poured into the cheese vat, and the starter culture is added. It is important that the starter culture is of adequate quality and is added in sufficient quantity to ensure rapid fermentation. The temperature of the milk should also be kept at an optimal level to ensure adequate starter culture activity. Starter culture activity may be measured by pH changes. The cheese vat and other equipment should be cleaned and disinfected before use. To prevent postpasteurization contamination, strict hygiene should be applied at this stage and later stages of production.
- *Rennet or rennet-like enzymes:* As for starter culture, it is important to obtain assurance from the supplier that the rennet is of adequate quality.
- *Setting the vat and cutting the curd:* The rennet or rennet-like enzymes are added and the milk is left undisturbed to form a gel. The coagulated milk is then cut into cubes. To prevent contamination with cutting utensils, hygienic practices are strictly observed.
- *Heating:* The curd is heated to 37–39 °C and the curd and whey are stirred. The curd shrinks as a result of heating and the whey is released. It is important to have control of temperature in order to maintain the activity of the starter culture. At the end of this step, the whey should have a titratable acidity of 0.14–0.16% and the curd pH should be approximately 6.
- *Draining and milling:* The whey is drained from the vat. The curd develops a plastic consistency as a result of continued acid development. The curd is cut into sections and passed through a mill to form ribbons. No major hazard is associated with this step. However, hygienic practices should be strictly observed to prevent recontamination.
- *Salting:* Salt is added to shrink the curd and further separate the whey. The amount of salt should be carefully controlled because it will have an impact on subsequent acid development and favor growth of *Staphylococcus aureus*. The salt content should be approximately 1.7%.
- *Pressing:* After salting, the curd is placed in a box and pressed. In this way, the remaining whey is removed. To prevent recontamination, this step should be carried out under strict hygienic conditions.
- *Aging:* The cheese is vacuum packed and aged at approximately 5 °C for a few weeks to few months. The control of temperature at this stage is important and critical for the prevention of growth of possibly present *Staph. aureus* and/or activity of decarboxylase-positive bacteria.
- *Storing and distribution:* To ensure safety and quality, storage should be at

refrigeration temperature and the product should be distributed rapidly.

RESEARCH NEEDS

Research needs in the area of fermentation are extensive and relate to several areas.

- identification of new organisms and technological developments leading to new products with enhanced nutritional or organoleptic quality
- assessment of safety of starter cultures, be they traditional organisms selected for their specific features or genetically modified organisms
- interaction of LAB with pathogens in the intestinal tract and risks for transfer of pathogenicity and virulence
- potential of fermentation technology to control pathogens
- identification and selection of organisms with potential health benefits
- transfer of technology to settings where the technology can be of specific benefit to health
- implementation and evaluation of health education interventions carried out to ensure safe application of fermentation.

Additional guidance on research needs and priorities in this area are provided elsewhere.[6,18]

CONCLUSION

Fermentation is and will continue to remain an important technology. For foods for infants and young children, it presents clear nutritional and safety advantages. In the industrialized countries, the technology may not be important as a preservation technique, but advances in areas such as molecular biology will help in the development of new products, with diversified taste and aroma and perhaps health benefits. The challenge will be to ensure that organisms used as starter culture are safe and that the process is designed and applied in such a way that potential hazards, in particular acid-resistant pathogens, are controlled.

The greatest benefits but also risks are currently in the developing countries and in places where the technology is used in an artisanal way. In the developing world, where refrigeration facilities are lacking, the technology can make a great contribution to public health by preventing the growth of pathogens. However, substantial health education campaigns are necessary to ensure that the technology is applied in an appropriate manner and leads to safe and nutritious products.

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