Storage Temperatures Necessary to Maintain Cheese Safety

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SUMMARY

Available information on bacterial pathogen growth, stasis, and death in cheeses was reviewed and evaluated to determine storage temperatures necessary to maintain product safety. In view of the variety and large volume of cheeses consumed throughout the world, the incidence of foodborne outbreaks associated with cheeses is extremely low. Research revealed that the inherent characteristics of most cheeses create a hostile environment for bacterial pathogens, especially at elevated ripening and storage temperatures. Therefore, it is recommended that the following cheeses, manufactured in the United States with pasteurized or heat treated (> 63°C for >16 seconds) milk, should be exempt from refrigeration requirements during ripening, storage, shipping, and display: Asiago (medium and old), Cheddar, Colby, Feta, Monterey Jack, Muenster, Parmesan, Pasteurized process, Provolone, Romano, and Swiss/Emmentaler. It must be stressed that the manufacture of these cheeses must be done under the proper conditions of Good Hygiene Practices, Good Manufacturing Practices, and HACCP principles, and according to CFR requirements. In addition, the natural cheeses must include active cultures, and the storage and display temperatures must not exceed 30°C.

INTRODUCTION

Temperature-dependent storage of most foods has three major roles – to allow for curing/ripening of foods that contain added active starter cultures and enzymes, to prevent quality defects, and to control pathogen growth. In making decisions on whether a food requires time/temperature control for safety, the properties of the food itself must be considered *(3)*. The role of temperature-dependent aging and storage is similar for cheese and for other foods, but the targets differ significantly because of unique inherent characteristics of the finished food product.

Transformation of chalky, acid-tasting curd into ductile, full-flavored cheese is accomplished during ripening through the action of milk enzymes, rennet, and various organisms in the cheese, including those in the starter culture. The biochemical changes that occur during cheese ripening are complex and involve fermentation of the carbohydrate; hydrolysis of fats and proteins with subsequent decarboxylation, deamination, and/or hydrogenation; and production of carbonyls, nitrogenous compounds, fatty acids, and sulfur compounds all of which contribute to the overall body, texture, and flavor of the final product *(63)*. These inherent characteristics also create a hostile environment for pathogens *(25)*. This review of scientific information on pathogen death and growth in cheeses at various storage temperatures will determine parameters necessary to ensure safety of cheeses in the marketplace. The United States cheese industry advocates the use of a science-based approach for assessing the risk posed by ready-to-eat foods for possible transmission of pathogens in the food supply *(24)*. Applying HACCP principles enhances the manufacture of safe cheese *(35)*.

In view of the variety and large volume of cheese consumed throughout the world, the incidence of outbreaks of food poisoning and foodborne disease associated with cheese are extremely low *(36)*. Epidemiology studies of cheese-related outbreaks in the United States, Canada, and Europe have found no outbreaks linked to hard Italian varieties, e.g., Parmesan, Romano, and Provolone. Varieties such as Cheddar and Swiss were infrequently involved *(38)*. In general, very few documented illness outbreaks have been linked to consumption of properly ripened hard cheese. Therefore, time/ temperature control of hard cheese is primarily needed not for safety reasons, but to maintain the organoleptic quality of cheese *(3)*.

INHERENT CHARACTERISTICS OF SAFE DAIRY FOODS

Numerous researchers have reported bactericidal and/or bacteriostatic effects on pathogenic bacteria in foods because of reduced moisture, low water activity, low pH as the result of organic acid production, salt, heat treatment, competing flora, biochemical metabolites, bacteriocins, and ripening, either singly or as part of hurdle technology *(1, 3, 5, 6, 10, 11, 13, 15, 17, 22, 25, 26, 29, 34, 36, 37, 38, 39, 40, 43, 45, 48, 49, 51, 58, 59, 64, 65, 66, 68, 69, 70, 76)*. Refrigeration cannot be depended upon to reduce the number of pathogens, as it has been proven that *Listeria monocytogenes* (*L. monocytogenes*) and other psychotropic pathogens are capable of growth at these temperatures. Therefore, other factors, such as diligence with regard to good hygiene practices by the food industry, must be responsible for the lack of pathogen growth in fermented dairy foods. Results also confirm the low frequency of contamination by *L. monocytogenes* of pasteurized fluid milk products sold in the United States *(24)*.

INHERENT CHARACTERISTICS OF CHEESE

Cheeses are one of the oldest types of prepared foods. Cheesemaking provided human kind with a means of concentrating and preserving milk at a time when refrigeration was unknown and principles of food preservation were vague empirical concepts at best *(52)*. The vast majority of cheese manufactured in the United States is made from pasteurized or heat-treated milk, which renders the product free of most pathogens *(38, 39, 40).* The inherent characteristics of cheeses made with starter culture addition provide multiple hurdles that inhibit pathogen growth *(3, 47)*. A multiplicity of practices other than pasteurization or heat-treatment also contribute significantly to the microbiological safety of cheese *(10, 11, 38)*. Some practices, such as milk quality management, lactic culture protocols, pH control, salt addition, and controlled curing conditions, are established technologies *(38)*. Other factors may include natural inhibitory substances (e.g., lysozyme), starter metabolites and fermentation by-products (e.g., nisin), including organic acids (e.g., lactic, acetic, propionic, and formic). Water activity/ moisture content imposes additional detrimental effects on foodborne pathogens during the manufacturing and ripening of cheese *(10, 11, 38, 66)*.

During the manufacture of semi-soft, hard, and very hard cheeses, the cheese is subjected to relatively long exposure to ideal incubation temperatures for bacteria. For example, Cheddar and related varieties are maintained at 31–39°C during manufacture and are formed or hooped at temperatures in the 32–37°C range. Many Cheddar-type cheeses are cured or aged at temperatures up to 15.6°C. Swiss cheese is held for a period of 4–8 weeks at a temperature of 22.2–23.3°C to develop the characteristic eyes and flavor. If storage of Cheddar and Swiss cheese at room temperature had any inherent detrimental effect on safety of these cheeses, then neither would be safe to consume *(51)*.

Specifically for *L. monocytogenes*, numerous studies suggest that the composition of cheese, ripening and storage conditions, lactic acid cultures, pH, salt, and moisture concentration influence its survival and growth *(15, 29, 39, 40, 43)*. The fate of *L. monocytogenes* and other foodborne pathogens during cheese ripening is determined by the microbiological, biochemical, and physical properties of the particular cheese *(43, 64)*. Thus, cheese is a very complex system, with the following factors acting simultaneously to determine the behavior of *L. monocytogenes* during ripening: (a) type, amount, and activity of starter culture; (b) pH as determined by concentrations of lactic, acetic, formic, and other acids; (c) presence of hydrogen peroxide, diacetyl, and various antimicrobial agents (Nisin, diplococcin, and other bacteriocins); (d) levels of nutrients, salt, moisture, and oxygen; and (e) the cheese ripening temperature *(64)*.

Fermentation is an age-old food preservation method used to inhibit the growth and survival of pathogenic bacteria *(48)*. Lactic acid bacteria commonly used to produce fermented dairy products are antagonistic to foodborne pathogens and will either inhibit their growth or inactivate them *(5, 13, 36, 59, 66, 70)*. In addition, research has shown that some starter cultures are detrimental to food spoilage organisms as well as various pathogens in these products *(1, 17, 22, 51, 58, 69, 76)*. Responsible for this action are metabolites such as lactic and other acids, diacetyl, hydrogen peroxide, and various antibiotic-like substances produced by lactic acid bacteria, which are probably synergistic *(34, 36, 37, 45, 49, 66)*.

Examples of pathogens that are susceptible to inactivation or growth inhibition by metabolites of lactic acid bacteria include *Salmonella* Typhimurium, enteropathogenic *Escherichia coli*, *Staphylococcus aureus,* and *L. monocytogenes (66)*. Growth of *L. monocytogenes* is always inhibited appreciably in lactic acid cultured product when compared to that of the control, no matter how high the final pH of the fermented milk. Even when the final pH dropped only to 5.99, growth of the pathogen was inhibited by 84% relative to the control *(65)*. This suggests that factors other than the hydrogen ion concentration are involved in the inhibition of *L. monocytogenes* by lactic acid bacteria *(65)*. These observations have been documented by other researchers, who noted that lactic cultures inhibited pathogens such as salmonellae and staphylococci, even when pH was controlled at 6.6 *(26)*. Modern lactic culture technology for cheese manufacturers has virtually eliminated *Staphylococcus*-caused outbreaks involving cheese *(40)*. Vigorous starter growth should protect fermented milk products against the growth of pathogens and the formation of staphylococcal enterotoxin *(36)*. Mathew and Ryser *(48)* reported increased injury of healthy *L. monocytogenes* cells during fermentation; at the end of the 24-h fermentation period, > 90% of the healthy *L. monocytogenes* cells were injured. Additionally, at the end of the product's shelf life, > 99% of the initial population was injured, and no significant decrease in the percentage of injury was observed. It was also discovered that the presence of *L. monocytogenes* did not adversely affect the growth of the starter culture at any inoculation level *(48)*. Gengeorgis et al. *(25)* demonstrated that non-soft cheeses made with the use of starter cultures and pH values of < 5.5, as well as processed cheeses, will not support growth of *L. monocytogenes* at 4 to 30°C if the cheeses are contaminated from raw foods after the consumers open packages. Rapid acid production is the principal factor responsible for the elimination of pathogens from semi-hard cheese. The use of an effective starter culture is not only critical for preventing growth of pathogens, but also essential for the production of good quality cheese *(6)*. The preservative effect of lactic acid bacteria can be attributed partly to the activation of the lactoperoxidase system and partly to bacteriocins *(4)*.

Temperatures of curd cooking and aging/curing/ripening/storage have an impact on pathogen growth and survival in cheese. In hard cheese types with higher curd cooking temperatures, growth is slight *(68)*. There is considerable evidence showing that certain cheeses do not support growth of pathogens during the aging process and subsequent storage *(11)*. A review of the literature related to the potential for growth of pathogens in hard cheeses that are aged for at least 60 days shows that such growth is not likely to occur because of factors inherent to these cheeses *(31)*. Pathogens that survive the manufacturing process decrease faster at higher storage temperatures *(14)*. The death rate of *Salmonella* in Samsoe cheese was slower at 10–12°C than at 16–20°C *(36)*. It has been concluded that, for traditionally made hard cheeses, time/temperature control for safety is not required *(3)*.

In most cheese varieties, salt concentrations attain levels of 1.6–3.0% of the total weight of the cheese, which would not affect most of the pathogenic bacteria in cheese. But it must be realized that salt is dissolved in the aqueous phase of the cheese only, the actual site of bacterial growth. Given the respective calculated values, salt concentrations in the aqueous phase reach levels of 2.2–6.5% or higher and will, in fact, at least slow down the growth rate of most bacteria and even have a detrimental effect on the more sensitive ones *(68)*.

Where scientific data do not exist, all the inherent characteristics of cheese can serve as criteria in determining potential growth of pathogens by the use of mathematical modeling *(16, 72, 79, 83)*. When two or more of these criteria are combined, the resultant effect is an additional hurdle to the outgrowth of pathogens of concern. It is this effect that makes it possible to store certain cheeses safely beyond either one of the two Food Code criteria for date marking and refrigeration (i.e., 7 days at 5°C or 4 days at 7.2°C). This led the US Food and Drug Administration to issue, on December 15, 1999 *(11)*, a letter suggesting that regulatory agencies use their discretionary authority and defer enforcement action regarding date marking aged hard cheeses. In that letter, FDA granted a formal interpretation to the Food Code that hard and semisoft aged cheeses and pasteurized process cheese, each manufactured according to 21 CFR 133 as specifically cited above and maintained under refrigeration, are exempt from the Food Code's date marking provision related to refrigerated, ready-to-eat, potentially hazardous food. This interpretation has subsequently been incorporated into state statutes, such as Wisconsin's *(2)*. Feta cheese was later added to this exemption list by FDA (in the case of Iowa Dept. Health vs. Shullsburg Creamery).

<u>The FDA/USDA evaluation classified cheeses as follows: Fresh soft – Queso fresco, Queso de Crema, Queso de Puna Soft unripened (> 50% moisture) – Cottage, cream, Ricotta Soft ripened (> 50% moisture) – Brie, Camembert, Feta, Mozzarella Semi-soft (>39–50% moisture) – Blue, Brick, Monterey Jack, Muenster, Provolone Hard (< 39% moisture) – Cheddar, Colby, Parmesan, Processed</u>

SPECIFIC CHEESES AND THEIR INHERENT CHARACTERISTICS

Cheeses are typically categorized according to their moisture content:

<u>Soft > 50% Semi-soft > 39 – < 50% Hard < 39% *(4, 22)*</u>

Hard and semi-soft cheeses are the focus of this research review.

Research by Gengeorgis and colleagues *(25)* has yielded results indicative of those obtained by other researchers, which prove death of pathogens in nonsoft cheeses stored at various temperatures. In this study, 49 market cheeses representing 24 varieties were purchased commercially. Cheeses were inoculated with 104 cells of *L. monocytogenes* per square cm. The inoculum was a cocktail of 5 strains — Scott A, V7, RM-1, VPH1, VPH2. Inoculated cheeses were stored at 4, 8 and 30°C for up to 36 hours. Certain cheeses (Queso Fresco, Panela Ranchero, Ricotta, Teleme, Brie, Camembert, and Cottage) supported *Listeria* growth in cheese at one of the storage temperatures. Cheeses not supporting growth but causing gradual death at all temperatures included Cotija, Cream, Blue, Cheddar, Monterey Jack, Swiss, Colby, String, Provolone, Muenster, Feta, and Kasseri with pH values of 4.3–5.6; Process Cheese (pH 5.7–6.4); and Limburger cheese (pH 7.2). Overall, this study demonstrated that nonsoft cheeses made with the use of starter cultures and at pH values of < 5.6, as well as processed cheeses, will not support growth of *L. monocytogenes* at 4–30°C if contaminated from raw foods (meat, poultry, fish, and vegetables) after the opening of the packages by consumers. In all cheeses that caused gradual death (Cotija, Cream, Blue, Cheddar, Monterey Jack, Swiss, Colby, Provolone, Muenster, Feta, Kasseri, Process, Limburger), death at 30°C was greater than or equal to death at 4°C.

Cheddar
Cheddar is a hard cheese that does not support *L. monocytogenes* growth and that causes gradual death at all temperatures *(25)*. This finding is confirmed by an FDA correspondence *(11)* and also agrees with work by Ryser and Marth *(61)*, who reported that growth of *L. monocytogenes* during Cheddar cheese manufacture appeared to be inhibited by proper acid development resulting from an active starter culture. Behavior of other pathogens during Cheddar manufacture and ripening show similar results. With normal starter activity, inoculated *Staphylococcus aureus* died rapidly *(60)*, as did *Yersinia enterocolitica (67)*. Norholt *(54)* illustrated die-off of *Salmonella* spp. After 2 weeks. Wood et al. *(84)* found that, of 11 vats of *Salmonella*-contaminated Cheddar cheese curd, only 2 remained positive in the finished cheese immediately after manufacture. In 1 and 4 months, these 2 vats were clear of the inoculated

Salmonella. This result is supported by studies of Goepfert et al. *(28)* and Hargrove et al. *(32)* in artificially inoculated Cheddar. Both groups found a 75– 80% reduction in *Salmonella* after hooping and pressing during manufacture.

Numerous researchers have reported kill of pathogens at higher ripening and storage temperatures. *Salmonella* spp. survived longer when Cheddar cheese was stored at 4.5°C rather than 10°C *(82)*. In general, a low pH and a high ripening temperature result in a higher inactivation rate for pathogenic organisms *(61)*. Using stirred-curd Cheddar cheese, Goepfert et al. *(28)* showed that the number of *S.* Typhimurium decreased by a factor of 10,000 during 10–12 weeks of ripening at 13°C, whereas a similar decrease required 14–16 weeks at 7.5°C. Park et al. *(58)* reported that salmonellae survived during ripening of Cheddar cheese for up to 7 months at 13°C and 10 months at 7°C. Ryser and Marth *(61)* reported an inactivation rate of *L. monocytogenes* 0.9 logs less at 6°C than at 13°C. International Dairy Federation researchers demonstrated that the decrease in numbers of staphylococci in Cheddar was greater at higher temperatures (10°Cand 13°C) than at 7°C *(36)*.

Colby

Colby is a hard to semi-soft cheese that does not support *L. monocytogenes* growth and causes gradual death at all temperatures *(25)*, a finding confirmed by an FDA correspondence *(11)*. Various researchers studying the behavior of inoculated pathogens during Colby cheese manufacture and ripening determined that *E. coli* generally decreased over a period of weeks and was not detected after 4–6 weeks *(41)* and that numbers of *Y. enterocolitica* generally decreased over a period of weeks at 3°C *(51)*. Yousef and Marth *(85)* found that, early in storage of Colby cheese, numbers of *Listeria* in the cheese remained relatively constant for a time that depended on the strain used. Numbers of *Listeria* in cheese decreased steadily thereafter at a rate that depended mainly on composition of the cheese. It should be noted that 2 of the 6 lots of cheese manufactured in this study had moisture levels higher than CFR specifications. IDF researchers demonstrated that the decrease in numbers of staphylococci in Colby was greater at the higher temperatures (10°C and 13°C) than at 7°C *(36)*.

Monterey Jack

Monterey Jack is a hard to semi-soft cheese which does not support *L. monocytogenes* growth and causes gradual death at all temperatures *(25)*. Other than this referenced study, there exists little published research with this cheese. However, it is very similar, with regard to pH, aqueous NaCl, and moisture, to other cheeses that have been heavily studied and proven not to support pathogen growth.

REGULATORY EVALUATION

In a series of correspondences, in a letter form as an inclusion to the US FDA Program Information Manual on retail Food Safety and in a subsequent correspondence *(11, 31)*, FDA exempted the following cheeses from the date marking mandate within the US Food Code:

Asiago Limburger
Blue Monterey Jack
Brick Muenster
Cheddar Parmesan
Colby (< 40% Pasteurized moisture) process
Edam Provolone
Feta Reggiano
Gorgonzola Romano
Gouda Sapsago
Gruyere Swiss/Emmentaler

In 2001, FDA/USDA *(77)* conducted a risk analysis of foodborne outbreaks of *L. monocytogenes* from ready-to-eat foods (Table 1). The evaluation revealed that there was a very low risk for listeriosis by Feta cheese, heat-treated natural and process cheeses, and aged cheeses *(77)*.

Obtaining more information from research, industry, and regulatory experience, FDA/USDA *(78)* updated their *L. monocytogenes* risk analysis in 2003 with the following results (Table 2). Utilizing a cluster analysis of predicted risk that takes into account the relative risk of listeriosis for the total population on a per serving and per annum basis, the following risk categories were developed for cheese:

- **High risk – soft unripened cheeses (cottage, cream)**
- **Moderate risk – fresh soft cheeses (Queso Fresco) soft ripened cheeses (Brie, Camembert, Feta, Mozzarella) semi-soft cheese (Blue, Brick, Monterey Jack)**
- **Very low risk – hard cheeses (Cheddar, Swiss, Parmesan)**
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Process cheeses

FDA/USDA actually decreased the predicted risk of soft ripened and certain semi-soft cheeses to “Moderate” due to increased use of pasteurized or otherwise heat-treated milk, and effective food safety control programs. The very low risk cheeses have similar characteristics of being subjected to bactericidal treatment, having very low contamination rates, and possessing an inherent characteristic (or two) that either inactivates *L. monocytogenes* (hard cheese) or prevents its growth (process cheese). As can be noted from this review, many more cheeses fit this category than recognized by USDA. The relative risk indices used may not give a clear picture of the range of risk potential that exists. The differential between per-serving risks associated with deli meats (relative risk rank of 1) and hard cheeses (relative risk rank of 23) is almost 10,000,000fold *(78)*.

CONCLUSIONS

Science-based data presented herein adequately illustrate the fact that most cheeses containing < 50% moisture (or more, in the case of Feta) and active lactic acid starter cultures, along with traditional levels of salt, pH, fat, etc., do not allow the growth of pathogens at temperatures between 4 and 30°C. In fact, in the vast majority of the cheeses, a higher temperature during ripening/aging and storage leads to significant bactericidal activity. A summary of the reviewed science and data is available in Table 3. Mathematical models were generated using the USDA Pathogen Modeling Program, but given that this system is in nutrient broth, not in a limited moisture solid food (cheese), growth/death curves generated were meaningless. No other models reviewed were found to be appropriate.

RECOMMENDATIONS

For cheeses manufactured in the United States with pasteurized or heat-treated (> 63°C for > 16 s) milk, under hygienic conditions outlined in Good Hygienic Practices, Good Manufacturing Practices, and HACCP systems, using active lactic acid cultures, and according to CFR specifications, the following cheese should be considered by regulatory agencies (FDA, USDA, state, local, etc.) exempt from any and all refrigeration requirements for aging, storage, shipping, and retail display, with a maximum temperature of 30°C:

Asiago (medium and old)
Cheddar
Colby
Feta
Monterey Jack
Muenster
Parmesan
Pasteurized process cheese
Provolone
Romano
Swiss / Emmentaler

If this exemption would apply only to pre-packaged cheeses, Parmesan and Romano, and possibly medium and old Asiago — because of their inherent characteristics — would not have to be prepackaged for this refrigeration exemption. Soft/fresh Asiago, Blue, Brick, cream and Mozzarella require further investigation before a recommendation for exemption could be made. **There is one common thread among all the ripened cheeses evaluated (this would exclude Mozzarella); the curing/ ripening/aging step is detrimental to bacterial pathogens, especially at elevated temperatures up to 30°C. Therefore, for safety purposes, refrigerated storage of the cheeses would appear to be unnecessary and possibly counterproductive.**

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