



## Reduction of pasteurization temperature leads to lower bacterial outgrowth in pasteurized fluid milk during refrigerated storage: A case study

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### ABSTRACT

Bacterial numbers over refrigerated shelf-life were enumerated in high-temperature, short-time (HTST) commercially pasteurized fluid milk for 15 mo before and 15 mo after reducing pasteurization temperature from 79.4°C (185°F) to 76.1°C (169°F). Total bacterial counts were measured in whole fat, 2% fat, and fat-free milk products on the day of processing as well as throughout refrigerated storage (6°C) at 7, 14, and 21 d postprocessing. Mean total bacterial counts were significantly lower immediately after processing as well as at 21 d postprocessing in samples pasteurized at 76.1°C versus samples pasteurized at 79.4°C. In addition to mean total bacterial counts, changes in bacterial numbers over time (i.e., bacterial growth) were analyzed and were lower during refrigerated storage of products pasteurized at the lower temperature. Lowering the pasteurization temperature for unflavored fluid milk processed in a commercial processing facility significantly reduced bacterial growth during refrigerated storage.

**Key words:** pasteurization temperature, milk, spore-forming bacteria

### INTRODUCTION

The public health objective of milk pasteurization, as defined in the 2009 Pasteurized Milk Ordinance (PMO; FDA, 2009), is to eliminate all non-spore-forming human pathogens commonly associated with raw milk. The PMO specifies heating milk to a minimum of 72°C for 15 s to achieve HTST pasteurization. In the absence of postpasteurization contamination (PPC), the microorganisms that limit fluid milk shelf-life are the spore-forming bacteria *Bacillus* spp. and *Paenibacillus* spp. (Fromm and Boor, 2004; Durak et al., 2006; Huck et al., 2007b, 2008). These organisms are found ubiquitously in different environments and can enter the raw milk supply at various points (Huck et al., 2008). Although

several spore-formers can survive HTST pasteurization in spore form (Collins, 1981; Huck et al., 2007a), not all can reproduce in fluid milk at refrigeration temperatures. For example, although many *Paenibacillus* spp. isolates obtained from milk processing systems can grow in milk at temperatures around 6°C, relatively few *Bacillus* spp. isolates are able to do so (Fromm and Boor, 2004; Ranieri and Boor, 2009). The shift in the microbial ecology of refrigerated pasteurized milk from predominantly *Bacillus* spp. in freshly pasteurized milk to *Paenibacillus* spp. after approximately 14 d (Fromm and Boor, 2004; Ranieri et al., 2009) implicates the presence of *Paenibacillus* spp. in pasteurized milk as an important cause of fluid milk spoilage in the absence of PPC.

It is common for HTST pasteurization to be performed above the minimum parameters stipulated by the PMO (Fromm and Boor, 2004; Huck et al., 2007b; Martin et al., 2011). Further, in response to food safety concerns raised in the early 2000s, many milk processors raised pasteurization temperatures well above minimal levels. However, several processors anecdotally reported increased numbers of consumer complaints about milk spoilage after pasteurization temperatures were increased (Ranieri et al., 2009). A controlled study designed to measure the effects of pasteurization temperatures on postpasteurization bacterial outgrowth showed that milk processed at higher HTST temperatures had greater bacterial outgrowth during refrigerated storage than milk processed at lower temperatures (Ranieri et al., 2009). In response to the Ranieri et al. (2009) study, a small-scale New York State dairy processor reduced its HTST pasteurization temperature from 79.4°C (185°F) to 76.1°C (169°F). In this report, we compare bacterial numbers in commercial fluid milk processed before and after the change in pasteurization temperature.

### MATERIALS AND METHODS

#### *Sample Processing and Plant Characteristics*

Raw milk processed at the small-scale, New York State licensed dairy processing facility was obtained

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from a single farm. Approximately 1.5 million lbs of raw milk are processed into fluid milk and other products each year at the facility.

Raw and pasteurized milk samples were collected from the processing facility once a week for 30 mo. Processing conditions during the first 15 mo were 79.4°C (185°F) for 18.25 s. During the second 15-mo period, processing conditions were 76.1°C (169°F) for 18.25 s. Pasteurized samples included (1) whole-fat milk packaged in half-gallon (1.89-L) plastic or 10-oz (295-mL) paperboard; (2) 2% milk packaged in half-gallon (1.89-L) plastic or 10-oz (295-mL) paperboard; and (3) skim milk packaged in half-gallon (1.89-L) plastic or 10-oz (295-mL) paperboard.

### **Microbiological Analysis of Raw and Pasteurized Milk**

Raw milk representing each processing day was obtained in 2-oz (59-mL) vials from the processing facility before pasteurization. Milk was evaluated for total bacteria count by spiral plating on SPC medium (Difco, BD Diagnostics, Franklin Lakes, NJ) according to *Standard Methods for the Examination of Dairy Products* (Frank and Yousef, 2004) and coliform bacteria count on Petrifilm Coliform Count plates according to the manufacturer's instructions (3M, St. Paul, MN). Petrifilm and SPC plates were incubated at 32°C for 24 and 48 h, respectively.

All samples of whole-fat, 2% fat, and fat-free pasteurized milk were obtained from the processing facility on the day of processing in original 10-oz (295-mL) paperboard and half-gallon (1.89-L) plastic containers and held in those original containers at 6°C throughout shelf-life testing. Samples were evaluated on the initial day as well as on d 7, 14, and 21 postprocessing for total aerobic bacterial count on SPC agar and coliform bacteria count as described above for raw milk.

### **Statistical Analysis**

All statistical analyses were performed in JMP (version 7.0, SAS Institute Inc., Cary, NC). Microbiological data were log-transformed before performing ANOVA on bacterial numbers by day of shelf-life. Analysis of variance was also performed on bacterial growth by day of shelf-life, which was calculated by subtracting the total bacterial counts on the initial day from the total bacterial counts on d 7, 14, and 21. Pearson  $\chi^2$  was used to determine if a significant difference existed between the counts of samples with less than the PMO limit of 20,000 cfu/mL before and after the temperature change

at each day of shelf-life. Results are presented as percentages for ease of interpretation.

## **RESULTS AND DISCUSSION**

Using a small, licensed fluid milk processing plant that already produced high-quality fluid milk products with rare instances of postpasteurization contamination, we tested the hypothesis that reduced HTST temperatures that are still above the legal temperature of 72°C can further improve product quality and shelf life. This hypothesis was based on previous laboratory studies (Ranieri et al., 2009) that showed that a reduction in HTST pasteurization temperatures leads to reduced microbial growth in fluid milk stored under refrigeration temperatures.

### **High-Quality Raw Milk in Combination with the Absence of Postprocessing Contamination Results in High-Quality Pasteurized Milk**

The plant used for this study used a high-quality raw milk supply. Total bacterial counts in raw milk throughout the study period were below the PMO limit of 300,000 cfu/mL for commingled milk (FDA, 2009) with a mean of 3,800 cfu/mL and a maximum of 150,000 cfu/mL. High-quality raw milk should have an SPC count <10,000 cfu/mL (Murphy and Boor, 2000), and of the 181 raw milk samples in this study, 75.7% had SPC counts <10,000 cfu/mL. The raw milk sampled in this study had a mean coliform count of 21 cfu/mL, with a maximum of 540 cfu/mL, consistent with a previous study, which showed that 77% of 855 raw milk producer bulk tank samples from individual farms in New York State had a coliform count <100 cfu/mL (Boor et al., 1998). Although the PMO does not specify a regulatory limit for coliform numbers in raw milk, producer bulk tank raw milk samples have been reported to range from 5 to 4,130 cfu/mL (Jayarao and Wang, 1999). Raw milk with a coliform count of <50 cfu/mL is generally considered high quality (Murphy and Boor, 2000), and among the 181 raw milk samples in this study, 75.7% had coliform counts <50 cfu/mL.

The PMO specifies that grade "A" pasteurized milk have a SPC count of <20,000 cfu/mL and a coliform count of  $\leq 10$  cfu/mL throughout product shelf-life (FDA, 2009). Throughout this study (i.e., before and after implementation of a reduced HTST temperature), the pasteurized milk produced by the plant that was used for the study was of high quality with evidence of limited postpasteurization contamination. The microbiological tests conducted on all 604 pasteurized milk samples in this study show that on the initial day of

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