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A Field Topic

Seasonal and Processing Influences on Bacterial Count of Raw and Processed Milk

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ABSTRACT

An increase in total counts of raw milk was detected during summer in comparison to winter. The higher summer temperature did not affect the numbers of coliforms and staphylococci. The microbial contamination of pasteurized milk did not change significantly during the year. All bacterial counts increased during the flow of milk through the various systems of the dairy plant before the pasteurizer. It was shown that pipe lines could cause contamination of milk, especially at the beginning of flow after the line was temporarily shut down. A correlation between numbers of coliforms and staphylococci was found for both raw and pasteurized milk.

Many studies have been devoted to the microbial population of raw milk. The total number of aerobic bacteria has been found to be in the range of 10^3-10^6/ml, the number of psychrotrophic bacteria between 10 and 10^5 and the number of coliform 10-10^5 (*8,15*).

Other researchers (*3,8,11*) have found the number and types of organisms to be influenced by the season of the year. Dempster (*4*) studied the spatial variations in the psychrotrophic microflora of milk plants.

Staphylococcus aureus was usually found in raw milk samples (*2,14*). About 70% of the milk supplied in bulk in Holland has been found to contain 800-1300 coagulase- and DNAase-positive staphylococci/ml (*2*).

Elliott et al. (*5*) surveyed the microbial population of pasteurized milk and showed that the microbial quality of raw milk affected the organoleptic properties of the final product. Hankin et al. (*7*) found that immediately following bottling, pasteurized milk contained 500-1000 bacteria/ml and that the coliform count was usually less than 10/ml. Sheikh and Luedecke (*16*) showed that 4.9% of pasteurized milk samples contained coagulase-positive *S. aureus*.

Cleaning procedures in dairy plants are often inefficient in removing all bacteria from the equipment (*10,13*). Kato et al. (*9*) found many viable cells on the walls of the bulk cooler immediately after automatic cleaning. Marshall and Appel (*12*) surveyed 12 modern fluid milk plants and concluded that relatively large numbers of bacteria are introduced from pipe lines, valves and filling machines into the pasteurized milk.

This paper deals mainly with changes in bacterial counts of milk during processing from raw milk to a finished packaged fluid milk product.

MATERIALS AND METHODS

Sampling

Milk samples were collected aseptically at various points of flow of milk in a commercial dairy plant. A schematic sketch of the plant is shown in Fig. 1. The sampling points were: (*a*) the road tanker immediately after its arrival at the plant, (*b*) the storage tank, before pasteurization, (*c*) the entrance to the clarifying centrifuge, (*d*) the exit of the clarifier, (*e*) after pasteurization at the exit of the flow diversion valve (FDV) before the cooler and (*f*) the final product packaged in plastic bags.

To obtain samples the milk line was momentarily opened at the above points. The first few liters of milk were discarded, then a sample of about 500 ml was poured aseptically into a sterile flask.

Samples were taken from every point 10 times at intervals of 5 to 7 days, on different days of the week, during winter (January - February) and 10 times during summer (August-September). Samples were held in ice water until all microbiological tests were carried out (not later than 2 h after sampling).

In a previous study (*6*), it was found that stagnant zones contribute to bacterial contamination of milk flowing in pipelines.

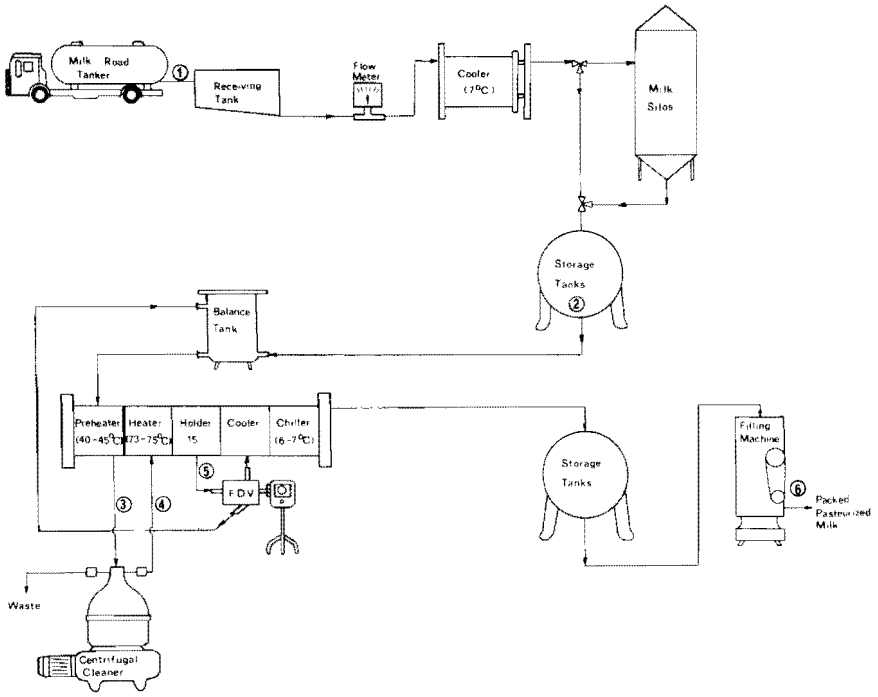

Figure 1. *Schematic description of pasteurized milk line and sampling points in it.*

Therefore milk from pipelines that were shut down for 24 h after automatic cleaning was also examined. Samples from the first milk and from the milk coming through after 10, 20, 30 and 40 min were collected.

Microbiological analyses

The number of aerobic bacteria, coliforms and staphylococci were determined in each milk sample. Standard media and incubation temperatures were used. Aerobic bacteria were counted on Plate Count Agar. The coliforms from raw milk were determined with Violet Red Bile Agar. After pasteurization, the coliforms were counted using the MPN method with enrichment in lactose broth and confirmation in Brilliant-Green Broth. Staphylococci from raw milk were determined directly with Baird-Parker (BP) plates. Staphylococci in samples of pasteurized milk were counted using the MPN method with Tryptic Soy broth + 8% NaCl as an enrichment medium. Bacteria from positive tubes were plated on BP. A few of the colonies from each BP plate were checked for the presence of coagulase enzyme.

RESULTS AND DISCUSSION

The average number of bacteria of the 10 samples taken in winter and the 10 samples taken in summer are given in Table 1. The total aerobic bacteria count was found to be in the same range as that observed by other investigators in different countries (*8,15*). However, the numbers of coliforms were slightly higher then those reported in the literature. On the other hand, the numbers of staphylococci found in this work were much lower than those found by Bijker et al. (*2*) in raw milk.

Since pasteurization reduced the number of bacteria by some orders of magnitude, statistical analysis of the results was carried out separately for raw milk and for pasteurized milk. The results of the analysis of variance are given in Table 2.

Raw milk

Two-way analysis of variance (*17*) showed that there was a significant difference between total counts in summer and in winter while the numbers of coliforms and staphylococci did not change significantly. In contrast to these results, it was found by Kielwein (*11*) and by Bogdanowicz and Mockiewich (*3*) that the numbers of coliforms change significantly between summer and winter.

For all three groups of bacteria tested there was a significant increase in numbers during the flow of milk in the plant up to the pasteurizer. To investigate which location contributed most to the increase in microorganisms, a Comparison of Contrasts was carried out (*17*). Results, for all three groups of bacteria, showed a significant increase in bacterial counts from the receiving point up to the pasteurizer at all the sampling points, except for the clarifier. This increase might have been caused by growth in stagnant zones or by a contribution of bacteria from the equipment. The clarifier did not reduce significantly the numbers of bacteria. Therefore, this unit which removes extraneous particles from milk did not affect microbial quality of the milk.

Pasteurized milk

Analysis of variance of the number of bacteria immediately after the FDV and the final product during the two seasons was carried out. The results showed that there was no significant difference between the number of bacteria in summer and in winter.

The increase in bacterial counts of the milk between the FDV and the final product was highly significant for coliforms and staphylococci. Anas (*1*) found that some recontamination of pasteurized milk occurred

TABLE 1. *Number of microorganisms in different stages[a] of the milk line.*

		Average number of microorganisms per ml					
		Before pasteurizer				After pasteurizer	
		Stage				Stage	
Group	Season	1	2	3	4	5	6
Total count	Winter	2.28×10^5	1.05×10^6	1.84×10^6	2.58×10^6	5.11×10^3	7.50×10^3
	Summer	2.96×10^5	1.84×10^6	3.74×10^6	4.52×10^6	4.54×10^3	5.98×10^3
Coliforms	Winter	1.10×10^4	3.94×10^4	5.71×10^4	3.91×10^4	0.311	3.94
	Summer	1.42×10^4	3.34×10^4	1.42×10^5	1.36×10^5	0.787	0.64
Staphylococci	Winter	3.11×10^2	7.02×10^2	1.28×10^3	1.61×10^3	0.385	1.07
	Summer	3.56×10^2	1.24×10^3	1.48×10^5	1.86×10^3	0.335	1.27

[a]Stages are shown in Figure 1.

TABLE 2. *Analysis of variance of the results obtained for raw and pasteurized milk.*

		Raw milk			Pasteurized milk		
Bacterial group	Source of Variation	Mean squares	Degrees of freedom	F value	Mean squares	Degrees of freedom	F values
Total count	Location	5.058	3	30.84***	0.205	1	0.45
	Winter/Summer	1.036	1	6.23**	0.056	1	0.12
	Interaction	0.033	3	0.20	0.006	1	0.01
	Error	0.164	72	—	0.459	36	—
Coliform	Location	2.984	3	5.39***	12.002	1	24.90***
	Winter/Summer	1.213	1	2.20	1.568	1	3.25
	Interaction	0.393	3	0.71	0.0005	1	0.01
	Error	0.554	72	—	0.482	36	—
Staphylococci	Location	2.003	3	10.80***	2.608	1	12.43***
	Winter/Summer	0.231	1	1.24	0.0005	1	0.002
	Interaction	0.034	3	0.23	0.046	1	0.22
	Error	0.185	72	—	0.210	36	—

** Significant at 97.5% confidence level.
***Significant at 99.0% confidence level.

in the regeneration and cooling sections of the plate pasteurizer. Other equipment units like storage tanks (*9*), pipe lines and filling equipment (*12*) could also contribute to the increase in bacterial load between the FDV and the final product. The increase of total count between the FDV and the final product was not significant. This could be explained by the fact that most bacteria appearing in the total count of pasteurized milk were not destroyed in the pasteurizer. Apparently conditions in pasteurized milk were not adequate to enable growth of these bacteria. In all instances, the phosphatase test after pasteurization was negative.

Contribution of pipelines to the microbial contamination

To evaluate the possible contamination of milk coming from pipelines, a line that was left idle for 24 h after automatic cleaning and sanitizing with chlorine was opened and the milk collected after different periods of time. This was done for different pipe lines. A typical result of such an experiment is shown in Fig. 2. It is clear that at the beginning of flow all the bacterial counts of the milk were higher by 1-2 orders of magnitude than after 30-40 min. This behavior was probably due to removal of bacteria from stagnant zones, as shown previously (*6*).

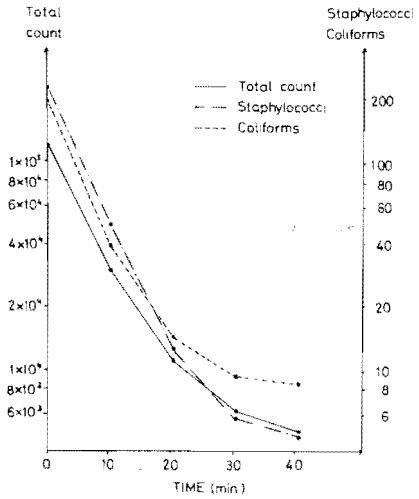

Figure 2. *Counts of microorganisms as influenced by time of flow of milk in a "clean" line.*

Relation between number of coliforms and staphylococci in the dairy plant

The correlation between numbers of coliforms and staphylococci inside the dairy plant was tested (120 pairs of data were compared). The results are given in Fig. 3. The regression coefficient (r) was 0.911, showing a significant correlation between these two groups in the plant. Since the origin of these two groups of organisms in milk is considered by some to be different, these results suggest that the sanitary conditions within the plant apparently control the numbers of these two groups.

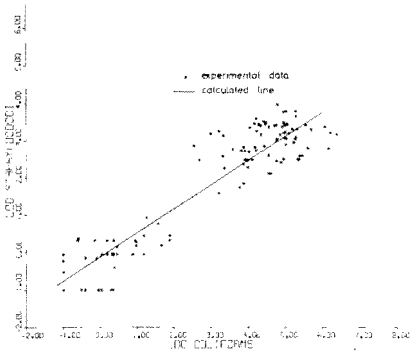

Figure 3. *Correlation between staphylococci and coliforms in milk.*

CONCLUSIONS

The higher ambient temperatures encountered during summer affected only the total count of raw milk but not the coliform and staphylococcus counts. This was probably due to the efficient cooling systems. Inside the dairy plant there was a significant increase of all types of bacteria examined, as the milk flowed through the equipment. Post-pasteurization contamination was observed.

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