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PSYCHROTROPHIC BACTERIA OF MILK COLLECTED IN A DAIRY FARM COOLING BASIN

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The psychrotrophic bacterial flora was examined in raw milk stored in dairy farm cooling basins for four milkings altogether. A standard counting method was used to determine the total count of psychrotrophs Petri dishes were incubated at 7±1^{0}C for 10 days (American Public Health Ass., 1978) and at 21^{0}C for 25 h (Oliveria and Parmelee, 1976). The total count of psychrotrophic bacteria in raw milk collected during four milkings ranged from six to seven log cfu/ml after incubation at 21^{0}C for 25 h and from five to seven log cfu/ml at 7^{0}C for 10 days. The results obtained show that the total counts of bacteria detected after incubation at 21^{0}C for 25 h were higher than those detected at 7^{0}C for 10 days. These differences amounted to one log cfu/ml in most of the raw milk samples tested.

Key words: psychrotrophic bacteria, raw milk

INTRODUCTION

In order to secure longer durability of food products, of both animal and vegetable origin, it became necessary to apply low temperatures on a large scale. Cooling devices at dairy farms, in transportation, within commerce as well as among consumers allowed the shelf-life of milk to be prolonged. However the storage of raw milk at cool temperatures for a long period of time creates a new problem with regard to the quality of raw milk (Cousin, 1982). Namely, some microorganisms are able to multiply at low temperatures, for which Eddy (1960) introduced the term psychrotroph. This term is used in the 13th edition of Standard Methods for the Examination of Dairy Products (American Public Health Ass., 1972) to indicate microorganisms which are able to form colonies in agar when incubated at 7^{0}C for 10 days. Psychrotrophic microorganisms contribute to the deterioration of raw milk quality by producing undesirable metabolites and enzymes (lipases and proteinases) among which many are thermoresistant (Adams, 1975; Gebre-Egziabher, 1980). Thus, they may cause spoilage of both raw milk and dairy products. Dependent on the extent of contamination by psychrotrophic bacteria, changes in the taste of raw milk (suety, rancid, bitter etc) may accur even before pasteurization is started. These unpleasant tastes are

reflected in the ultimate quality of products obtained from such pasteurized milk as well.

Our study was aimed at an examination of the effect of keeping raw milk in the dairy farm cooling basin for two-days on the growth of psychrotrophs. The parcipation of psychrotrophs in the total bacterial flora of raw milk was also determined. The number of psychrotrophic bacteria found in raw milk after incubation at 21^{0}C for 25 h (Oliveria and Parmelee, 1976) was compared with that obtained using standard counting methods for psychrotrophs (71^{0}C for 10 days).

MATERIAL AND METHODS

Raw milk from four milkings was collected into a cooling basin and examined on a trial farm in 1992. Samples of cooled raw milk were also taken after every single milking, that is after 12, 18 and 24 hours. Raw milk was cooled down to 6^{0}C. A total of 14 raw milk samples were analyzed altogether.

In the raw milk samples under test total counts of the mesophilic bacteria were made (30^{0}C for 72h) using methods referred to in "Pravilnik o metodama vršenja mikrobioloških analiza i superanaliza životnih namirnica" ("Sl. list SFRJ" No. 25/80).

For the determination of the total count of psychrotrophic bacteria a standard on-plate counting method was used in which Petri dishes were incubated at 7$\pm$1^{0}C for 10 days (American Public Health Ass., 1978) and at 21^{0}C for 25 h (Oliveria and Parmelee, 1976).

RESULTS AND DISCUSSION

Table 1 shows the contamination by mesophilic and psychrotrophic bacteria of raw milk stored in the cooling basins for four milkings.

The total count of mesophilic and psychrotrophic bacteria at the beginning of the trial indicated high initial contamination of raw milk. According to the official regulations ("Sl. list SFRJ" No. 45/83), all the samples of raw milk examined were bacteriologically unwholesome with regard to the count of mesophilic bacteria (>3x10^6/ml).

Growth of mesophilic bacteria during storage of the raw milk in the cooling basins was slow as the final count was only 44,18% above the initial count of mesophils in the first trial. This indicates that the cooling process throughout the storage-time was relatively effective. However, this did not apply to the second trial where the growth of these bacteria was rapid, so that at the end of the storage time numbers had increased by nearly one log cfu/ml (from six to seven log cfu/ml).

Lück (1972) found that newly expelled raw milk countained 50 psychrotrophs/ml only, but the count went considerably higher during manipula-

tion. According to Thomas and Thomas (1973) the proportion of psychrotrophic bacteria in the total count of the mesophils in hygienically produced raw milk is less than 10%, whereas in raw milk produced in unhygienic conditions it might reach more than 75%. La Grane and Nelson (1961) maintained that the count of psychrotrophs might frequently surpass the total count of mesophils in raw milk if it was obtained under unhygienic production conditions.

Table 1. Contamination of raw milk stored in dairy farm cooling basins during four milkings

Milking	Log 10 CFU/ml			Proportion of psychrophs in mesophilic bacterial flora (%)	
	Mesophils	Psychrotrophs			
	30°C; 72h	21°C;25h	7°C for 10 days	21°C;25h	7°C for 10 days
Trial 1					
I	6,63	6,46	5,89	60,42	16,04
I (after 12h of cooling)	6,62	6,43	5,72	64,28	12,38
I + II	6,59	6,46	5,71	74,35	13,08
I + II (after 18h of cooling)	6,49	6,41	5,72	83,87	16,77
I + II + III	6,53	6,46	5,86	85,29	21,47
I+II+III (after 24h of cooling)	6,82	6,57	6,34	56,06	33,33
I + II + III + IV	6,79	6,63	6,38	69,35	38,70
Trial 2					
I	6,53	7,04	6,62	323,50	123,50
I (after 12h of cooling)	6,64	7,04	6,83	250,00	154,54
I +II	6,63	7,00	6,74	232,56	127,90
I+II (after 18h of cooling)	7,18	7,11	7,04	86,67	73,33
I + II + III	7,23	7,20	7,04	94,12	64,70
I+II+III (after 24h of coolinhg)	7,23	7,11	6,64	76,47	25,88
I + II + III + IV	7,25	7,18	6,89	83,33	42,77

The results obtained here indicate that the raw milk was produced under unhygienic production conditions because the proportion of psychrotrophic bacteria in the total bacterial flora was 56,06 - 85,29% (21°C for 25 h) and 12,38 - 38,70 % (7°C for 10 days) in the first trial whereas in the second trial it was considerably higher than the total count of mesophils. Even at the end of storage it accounted for 83,33 % (21°C for 25h) or 42,77 % (7°C for 10 days) respectively. In this work, 286 colonies of Gram-negative and Gram-positive bacteria were isolated from raw milk and studied.

Some authors (Oliveria and Parmelee, 1976; Grifiths et al., 1980) believe that the time necessary for incubation of psychrotrophic bacteria might be reduced by using an incubation temperature near to the optimal temperature needed for growth of the majority of psychrotrophs. Oliveria and Parmelee obtained a high degree of correlation between the counts of psychrotrophic

bacteria detected at 21^{0}C for 25h and 7^{0}C for 10 days (r=0,992), whereas Grifiths et al., demonstrated it between 21^{0}C for 25h and 6^{0}C for 14 days (r=0,97).

After comparing the results for the total count of psychrotrophic bacteria at the temperatures used here, we concluded that the values obtained at 21^{0}C for 25h were higher than those obtained at 7^{0}C for 10 days. These differences amounted to one log cfu/ml in the majority of samples examined within the first and the second trial. These results confirm the references listed by Lawton and Nelson (1954) who showed that the growth of psychrotrophs was higher at 21 or 25^{0}C than at 5 or 10^{0}C. Bauman and Reinbold (1963) presented evidence that the highest count of psychrotrophs could be achieved at the incubation temperature of 20^{0}C. Piton and Ranvaux - Gaida (1990) considered that the exactness of a rapid method for determining psychrotrophic microorganisms (21^{0}C for 48h) was influenced by the nature of the microorganisms.

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PSIHROTROFNE BAKTERIJE MLEKA U RASHLADNIM BAZENIMA FARMI

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SADRŽAJ

Ispitivana je psihrotrofna mikroflora sirovog mleka čuvanog u rashladnim bazenima farmi tokom četiri muže. Za određivanje ukupnog broja psihrotrofa korišćen je standardni metod brojanja na ploči gde su Petri šolje inkubirane na 7± 1^{0}C za 10 dana (American Public Health Ass., 1978) i na 21^{0}C za 25h (Oliveria and Parmelee, 1976). Ukupan broj psihrotrofnih bakterija u mleku tokom četiri muže kretao se od šest do sedam log broja kolonija/ml kada je brojanje vršeno inkubacijom ploča na 21^{0}C za 25h i od pet do sedam log broja kolonija/ml na 7^{0}C za 10 dana. Rezultati ukupnog broja psihrotrofnih bakterija dobijenih inkubacijom ploča na 21^{0}C za 25h bili su veći u odnosu na 7^{0}C za 10 dana. Ove razlike su iznosile jedan logaritamski stepen kod većine ispitivanih uzoraka sirovog mleka.