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Microbiological and Physicochemical Properties of Raw Milk Used for Processing Pasteurized Milk in Blue Nile Dairy Company (Sudan)

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Abstract: An analysis of raw milk samples and pasteurized milk, after processing and before packaging demonstrated an association between the properties of the raw milk used and the pasteurized milk produced. Raw milk used provided by Blue Nile Dairy and Kordi farms. Raw milk from both farms was usually mixed and standardized (3- 3.2% fat) and pasteurized using High Temperature Short Time (HTST) pasteurizer. Raw and pasteurized milk were analyzed for chemical, physical and microbial properties. The result of raw milk for protein, lactose, sold not fat, ash, Density, freezing point, titratable acidity, and pH revealed significant variation ($P<0.01$) due to the source, while, fat was not significantly ($P>0.05$) affected by source. The mean values of microbiological results indicated that the log of cfu/ml of total bacterial counts (TBC), coliform counts, thermoduric bacterial counts and psychrotrophic counts were 4.755±0.007 and 1.294±0.021, 4.106±0.008 and 0.214±0.047, 2.980±0.014 and 0.621±0.049, 0.814±0.014 and 0.360±0.073 in raw milk samples and pasteurized milk at processing, respectively. The present study recommended that quality assurance programs should be started to ensure good quality milk and milk products.

Key words: raw, pasteurized, milk quality, microbiology, physiochemical properties, Sudan

INTRODUCTION

Milk is a highly nutritious food, ideal for microbial growth and the fresh milk easily deteriorates to become unsuitable for processing and human consumption (FAO 2001). High bacterial counts are indicator of poor production hygiene or ineffective pasteurization of milk (Harding 1999). Milk and milk products derived from dairy cows milk can harbor a variety of microorganisms and can be important sources of foodborne pathogens (Oliver *et al.* 2005; Yagoub *et al.* 2005). The presence of food-borne pathogens in milk is due to direct contact with contaminated sources in the dairy farm environment and to excretion from the udder of an infected animal (El Zubeir *et al.* 2006).

The hygienic quality problems of milk may arise from raw milk of diseased animals (Murphy and Boor 2000). Kang *et al.* (2005) reported that the presence of antimicrobial substances in raw milk could have serious toxicological and technical consequences. Raw milk may contain over 2,000,000 cfu/ml before processing of liquid milk or cheese making (Kameni *et al.* 2002).

The raw milk distributed for consumption in Sudan does not find the real quality control measures needed to be of good quality food (Mohamed and El Zubeir 2007). However, some new private dairy plants started the processing of fluid milk and some dairy products. These are faced with many problems of which the quality control measures constitute an important concern. Hence, the present study was designed to assess the chemical, physical and microbial properties of raw milk supplied to the Blue Nile Dairy Company plant (CAPO) and to compare it with the produced pasteurized milk.

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MATERIALS AND METHODS

Source of Milk:

This study was carried out during June to September 2005, and the raw milk for this study was collected from two dairies, namely Blue Nile Dairy Company and Kordi Farms. Blue Nile Dairy Company plant deals with both suppliers as a source of raw milk for processing pasteurized milk. Raw milk from both dairy farms is usually mixed before processing. Milk fat was standardized to 3-3.2%, and the milk was pasteurized at 72-76˚ C for 15 second using a high temperature short time (HTST) plate heat exchanger (Wincantor Pasteurizer, Wincanton Engineering Ltd, South Street Sherborne Dorset, U.K.). The pasteurized milk was packed into Tetra Pack container (Tetra Pack Technical Services AB, Ruben Rausing gata, 5-221 86 Lund, Sweden).

Raw milk samples (36 samples) and pasteurized milk after processing and before packaging (12 samples) were examined for total bacterial counts (TBC), coliform, psychrotrophic (PC) and thermoduric bacterial counts. Physiochemical properties **(**fat, protein, lactose, ash, solid not fat, density, acidity, pH and freezing point), antibiotics and phosphatase test were also done.

Chemical Analysis of Milk Samples:

The milk constituents (fat, protein, lactose, ash and solid not fat) and physical characteristics (density and freezing point), of the milk samples were determined by milk analyzer Lactoscan 90 (Aple Industries services–La Roche Sur Foron, France), according to manufacturer's instructions. Milk samples were mixed gently 4-5 times to avoid any air enclosure in the milk. Then 25 ml samples were taken in the sample-tube and put in the sample- holder one at a time with the analyzer in the recess position. Then when the starting button activated, the analyzer sucks the milk, makes the measurements, returns the milk in the sample-tube and the digital indicator (IED display) shows the specified results.

Antibiotic residues were determined using Delvotest® SP- ampule Kit (202– Delvotest SP 100, test box, DSM Food Specialties, the Netherlands). The method was carried out according to the manufacturer's instructions. Phosphatase test was done using Lactognost tables and powders obtained from Heyl, Chem. Pharm-Fabrik, 14167 Berlin. The procedure for phosphatase test was done according to the manufacturer's instructions. The acidity of the samples was determined according to AOAC (1990). The temperature and the pH of the samples were determined using pH– meter (Wagtech, HI 8314 membrane pH Meter, U.K.).

Microbiological Analysis:

The samples were examined for TBC, coliforms, thermoduric and psychrotrophic counts according to Houghtby *et al*. (1992); Christen *et al*. (1992); Ballou *et al*. (1995); Ravanis and Lewis (1995), respectively. Plate count agar No. 298 (Biomark Laboratories) was used for enumeration of TBC, thermoduric bacteria and psychrotrophic counts, while violet red bile agar No. 779 (Biomark Laboratories) was used to determine coliform counts. The media were prepared according to manufacturer's instructions. Plates for enumeration of TBC, thermoduric bacteria and coliforms were incubated at 32° C for 48 hours, 37° C for 48 hours and 37° C for 24 hours, respectively. Plates for enumeration of psychrotrophic counts were incubated at 7° C for ten days. Developed colonies were counted using manual colony counter. The plates counting 25-250 colonies were selected as described by Houghtby *et al*. (1992). The number reciprocal of the dilution factor was recorded as colony forming unit per ml (cfu/ ml). The milk ring test (MRT) for brucellosis was carried out according to Harrigan and McCance (1976).

Statistical Analysis:

Data were analyzed by SPSS programme (Statistical Package for Social Science, version 10.00). This test combines ANOVA with comparison of differences between means of the treatments at the significance level of $P< 0.05$.

RESULTS AND DISCUSSION

The means of fat, protein, lactose, ash and solids not fat content were 4.14%, 3.48 %, 4.33%, 0.778% and 8.58% in raw milk samples mixture (Table 1). The density, freezing point, titratable acidty and pH revealed 1.031, -0.520, 0.145 and 7.02 (Table 1). The analysis of variance showed highly significant variations ($P< 0.01$) due to the source of raw milk samples, except for fat. The composition of raw milks in the present study was compared favorably with the composition of milk in northern Europe, which contained fat of 4.3%, total protein of 3.4%, lactose of 4.65%, ash of 0.73%, TS of 13.3% and SNF of 9.0% (Invensys APV 2002). This

result also agrees with that reported by El Zubeir *et al.* (2005) for raw milk. The present study revealed lower mean values for lactose (%) than that reported by El Zubeir *et al.* (2005). The lower lactose may be due to the effect of psychotrophic bacteria (Ballou *et al*. 1995).

The results of physicochemical analysis of mixed raw milk used for producing fluid milk and the pasteurized milk. These results were higher compared to that reported by Elmagli and El Zubeir (2006a). These differences in milk composition may be due to initial raw milk used and the procedure of processing. However the results of freezing point agreed with those reported by Tetra Pak Processing Systems (2003) for freezing points of raw and pasteurized milk -0.520± 0.001 and -0.447± 0.000, respectively obtained during the present study (Table 1). This study also agreed with that reported by Elmagli and El Zubeir (2006a) who found the freezing point was -0.4734± 0.05032° C. The obtained data for acidity of raw and pasteurized milk of 0.145% and 0.143%, respectively, which are in line with that reported by Harding (1999), while the mean value was lower than that reported by Mohamed and El Zubeir (2007).

The microbiological quality of the raw milk used for processing pasteurized milk showed that the initial quality was good for TBC (log 4.800 cfu\ml), coliform counts (log 4.157 cfu/ml), thermoduric bacterial counts (log 2.994) and psychotrophic bacterial counts (log 810 cfu/ml) as shown in Table 1. The analysis of variance showed highly significant differences ($P< 0.01$) due to the source of raw milk samples for TBC and coliform counts. This result was lower than that reported by PMO (2001) for the average standard plate counts for can and bulk milk (700.000 bacteria /ml and 100.0 bacteria /ml, respectively). Moreover, the microbial standards for grade A raw milk is 100.0 bacteria/ml (PMO, 2001). The lower counts of bacteria may be due to good cleaning system and good handling from farms to the plant as was stated before by Chye *et al*. (2004).

Lower TBC value was obtained for pasteurized milk than that reported by Elmagli and El Zubeir (2006b), who reported a range of 6.5×10^5 to 6.5×10^{14}, but was similar to that of Reena *et al.* (2003). In addition, PMO (2001) reported that the bacteria standards for grade A pasteurized milk should be less than 20,000 bacteria /ml. Coliform bacteria counts of pasteurized milk showed lower numbers than these reported by Elmagli and El Zubeir (2006b). The lower coliform counts might be due to hygienic quality of raw milk, proper pasteurization process, good packaging and good storage conditions. This agreed with PMO (2001) who reported that the total bacterial standards for grade A pasteurized milk should be < 10 coliform/ ml. In addition, coliform counts obtained are in line with Sudanese Standards (SSMO, 2005) which stated that the maximum coliform counts should not to exceed 10^2 cfu/ml.

Table 1: Physical, chemical and microbial properties of raw milk to be used for processing pasteurized milk

Parameters	Mean ± S. E			
	Kordi Farm	CAPO Farm	Mixture	Pasteurized milk
Fat (%)	4.41±0.022	3.92±0.024	4.14±0.023	3.34±0.003
Protein (%)	3.47±0.012	3.42±0.014	3.48±0.013	3.21±0.004
Lactose (%)	4.32±0.022	4.25±0.024	4.33±0.024	4.00±0.005
Ash (%)	0.779±0.003	0.765±0.003	0.778±0.003	0.718±0.001
Solid not fat (%)	8.57±0.033	8.43±0.036	8.58±0.035	7.93±0.007
Density	1.030±0.106	1.030±0.117	1.031±0.114	1.028±0.003
Freezing point (° C)	-0.519±0.001	-0.513±0.001	-0.520±0.001	-0.447±0.000
Titratable acidity (%)	0.154±0.000	0.135±0.000	0.145±0.000	0.143±0.000
pH	6.91±0.003	7.04±0.003	7.02±0.003	7.06±0.001
Log TBC (cfu/ml)	4.700±0.013	4.700±0.013	4.800±0.014	1.294±0.021
Log coliform bacteria (cfu/ml)	3.935±0.014	4.227±0.015	4.157±0.015	0.214±0.047
Log thermoduric bacteria (cfu/ml)	2.994±0.024	2.953±0.026	2.994±0.025	0.621±0.049
Log psychotropic bacteria (cfu/ml)	0.789±0.023	0.843±0.026	0.810±0.025	0.360±0.073

TBC = Total bacteria counts

Table 2: Numbers and frequencies of milk ring test, antibiotic residues and phosphatase in milk samples at CAPO dairy factory

Measurement	Number of samples	Test			
		Milk ring		Antibiotic	Phosphatase
		+ve	-ve	-ve	-ve
Raw milk Kordi farm	12	12 (100%)	0	12 (100%)	ND
Raw milk CAPO farm	12	10 (83.33%)	2 (16.67%)	12 (100%)	ND
Raw milk mixture	12	12 (100%)	0	12 (100%)	ND
Pasteurized milk	12	0	12 (100%)	12 (100%)	12 (100%)

+ve = Positive, -ve = Negative, ND = Not done

Thermoduric bacterial counts (log 0.621cfu/ml) was lower than that reported by Mohamed and El Zubeir (2007). However, the present findings agreed with that reported by Invensys APV (2002) who reported an aerobic spores-forming bacteria of <400. The mean value of psychrotrophic bacteria for pasteurized milk was log 0.360 cfu/ml, which was lower counts compared with that reported by Elmagli and El Zubeir (2006b), who found the psychotrophic bacterial counts were <6.5×10 for pasteurized milk.

All the samples during storage showed the absence of the phosphatase test (Table 2). This result might be due to proper pasteurization. However, Elmagli and El Zubeir (2006a) demonstrated that 10 % of the pasteurized milk samples were positive to the phosphatase test.

No brucella antibodies were detected in pasteurized milk (Table 2), this might be due to proper pasteurization, and is in accord with that reported by OIE (2005). Moreover this result is better result than that reported by Alves *et al*. (2001). The presence of positive antibodies for brucella in the raw milk samples might suggest infection and/or vaccination, as those herds followed regular vaccination programmes. Similarly negative results of antibiotic residues test were obtained (Table 3), this may be due to proper follow up of antibiotic withdrawal periods which indicated the good quality of raw milk used. These results are in agreement with Van Schaik *et al*. (2002) and Yamaki *et al*. (2004).

It is concluded that the values of chemical contents are within standards limits except for lactose, whose value was lower than the reported by the dairy plant. Low TBC for pasteurized milk was obtained, and the results of this study clearly illustrate that pasteurization plays an important role in the survival and destruction of different bacterial contaminants.

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