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Quality Assurance of Marketed Raw Milk in Bhopal city, MP, India

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Abstract

The study was conducted to evaluate physicochemical and bacteriological quality of raw milk in Bhopal city (M.P) India. Milk samples were collected randomly encompassing whole city from milk vendors and milk vending shops for two consecutive years i.e. 2009-2010. The physicochemical parameters include Temperature, pH, Titrable acidity, Density and Conductivity, Additionally microbiological evaluation were based on Standard plate Count and Coliform counts. The results for milk vendors showed Temperature 18.19^oC, pH 6.61, Titrable acidity 0.183%, density 1.024g/ml, Conductivity 3.39 ms/cm and SPC 6.13 \pm 0.083 Ccfu/ml and Coliform count 4.01 \pm 0.1413 cfu/ml. furthermore the values of sample for milk vending shops were Temperature 13.6^oC, pH 6.34, Titrable acidity 0.181%, Density 1.026 g/ml, Conductivity 3.64 ms/cm, and SPC 6.37 \pm 0.085 Ccfu/ml and Coliform count 4.01 \pm 0.180cfu/ml. statistical analysis of data revealed that there is significant difference between sample of different sources at 5% significance level.

Keywords: Raw milk, physicochemical parameters, standard plate count, coliform count.

Introduction

Mankind has been respecting milk as divine drop of health since the dawn of civilization. It is considered as an excellent culture medium for multiplication of several bacteria. They grow and multiply in milk and causes chemical changes that make it unpalatable. Quality of the raw milk can be judged by microbial load. As per the guide lines of International Dairy Federation the production of milk having Standard Plate Count of 10^4 Cfu/ml reflects good hygienic practices while high initial Standard Plate Count of more than 10⁵ Cfu/ml are evidence of serious faults in production hygiene¹. Presently, India is the world largest producer of milk. Milk production in India is projected to be between 180-200 million tones by 2021-22 and the production of milk is expected to increase at the rate of 5% per annum². Only in Bhopal city thirty five thousand liters of raw milk is sold by milk vendors and milk vending shops. Raw unpackaged and unprocessed milk sold is much below standards in hygiene, their bacteriological quality needs to be rigorously monitored with regard to both number and type of micro flora present.

Material and Methods

Bhopal, the capital city is located in the state Madhya Pradesh in the central part of India. It is situated on 23^0 16' North latitude and 77^0 25' East longitudes and is located on a hard pink- red sand stone of Vindhya region.

Collection of samples: In this study sample of raw milk was collected randomly by selecting the shops and residential area from the different zones of Bhopal city. A total of 05 samples in triplicate from local vendors (those delivering milk on vehicles) and milk vending shops of each zone was collected for raw

milk. Multiple sampling was also done. Each sample was of 200 ml. for raw milk. These samples were collected for two consecutive years, January 2009 to December 2010. Samples of raw milk were collected in sterile glass bottles after proper mixing. Each sample container was sealed air tight after filling with sample. Containers were marked with source of sample, the date and time of sampling and other relevant information. After collection the samples were cooled and carried in ice box maintaining at a temperature of $4 - 5^{\circ}C$ and brought to the laboratory. They were preserved in refrigerators until analyzed.

Physico-chemical parameters: parameters analyzed in this study are Temperature, pH, Titrable acidity, Density and conductivity for the samples of raw milk as per the methods of AOAC³. Conductivity was measured by electro conductivity cell (Systronic, 304) according to the manufactures instruction. Prior to use it is calibrated using potassium chloride (KCl) reference solution. pH value of milk was determined by using a digital pH meter (Labatronics EI).

Bacteriological parameters- Samples of raw milk were examined for their Standard plate counts and Coliform counts⁴, ⁵. Ten fold serial dilutions of samples were made upto 10⁻⁸ in phosphate buffer. Triplicates of samples were plated using pour plate technique on Plate Count Agar Media (Himedia, M091) for Standard plate count and Violet Red Bile Agar (Hi media, M049) for Coliform count, petri plates were thoroughly mixed and allow to solidify then plates were incubated in inverted position at 37⁰C for 24 hrs after incubation plates containing colonies between 30 -300 were counted and results were expressed as Cfu/ml.

Statistical analysis: Number of bacteria present in 1 ml of

samples was calculated as per the formula^{6,7}. All microbial counts were changed to the \log_{10} of the number of colony forming units per ml (log 10 Cfu/ml) and from these Maximum, Minimum, Mean values and their Standard error were calculated. Data were analyzed using Analysis of Variance (ANOVA) and mean variation were compared by using Fisher's Least Square Deviation (LSD) to further interpret the results at 5% and 1% significance level by software (SYSTAT Version 13, 2011).

Results and Discussion

Mean annual temperature of samples collected from milk vendors ranged between 14.4° C to 23.2° C with a mean of 18.19° C ± 0.1420, for milk vending shops it varied from 13.6° C to 22.4° C with a mean of 18.03° C ± 0.4315 (figure -1). The mean temperature recorded from both sources (MV+MVS) in this study was almost similar (18° C), which is favorable for the growth of bacteria. Some variations in the mean temperature were recorded that depended on environmental fluctuations. Such an higher recorded temperature were indicative of lack of proper cooling during storage and distribution of raw milk samples and indicative of higher count of bacteria as recorded in Iran and Kenya^{8,9}.



pH of samples collected from milk vendors were in the range of 6.32 to 6.91 with a mean of 6.61 ± 0.0315 and for milk vending shops it ranged between 6.34 to 7.06 with a mean of 6.67 ± 0.0375 (figure-2). pH of the milk samples is related to acidity. Lower pH is indicative of higher developed acidities due to action of microbes where as higher pH values were obtained when cattle's were suffering from Mastitis^{10, 11}. In other studies various range of pH readings such as 6.44 - 6.99 have been reported^{12,13}.

Titrable acidity is a sum of natural and developed acidity and is an indication of quality of raw milk. According to Indian Standard the natural acidity of raw milk is 0.13% to 0.15%lactic acid¹⁴. Titrable acidity for the samples of raw milk collected from milk vendors ranged between 0.142 - 0.231%with a mean of $0.183 \pm 0.0044\%$.



In case of milk vending shops it ranges from 0.150 to 0.220 % with a mean of 0.181 \pm 0.0033 % lactic acid (figure-3). The difference in acidity was metabolic activities of microbes¹⁵. Longer period of storage is also responsible for higher Titrable acidity which in turn affects bacteriological quality of raw milk samples¹⁶.



Mean monthly variation in Titrable acidity

Density of raw milk samples collected from milk vendors were ranged between 1.015 -1.034 with a mean of 1.024 ± 0.0011 g/ml. Density of raw milk samples of milk vending shops varied from 1.017 to 1.033 with a mean 1.026 ± 0.0010 g/ml (figure-4). On average density of fresh whole milk is 1.029 g per ml¹⁷. Fat, a constituent of milk is responsible for its density as it is a lightest constituent of milk; Density is also lowered by addition of water¹⁸. It was noted that Tasci, recorded density 1.016 to 1.034 g/ml in her studies at Burdar (Turkey)¹⁹.





Mean monthly variation in Density

Conductivity is the ability of ionic matter to carry electric current. Conductivity of raw milk samples collected from milk vendors varied from 3.29- 3.48 mS/cm with a mean of 3.39 ± 0.0090 mS/cm. In the case milk vending shops it varied from 3.18 to 4.32 mS/cm with a mean of 3.64 ± 0.0600 mS/cm (figure-5). Mabrook and Petty studied the effect of milk composition on its electrical conductance they have reported that the mineral salts dissociated in the aqueous of milk played the dominant role in controlling this property; they have also reported that the conductance is affected by milk fat¹⁹. The samples of raw milk in the present study have exhibited lower conductivities. Such lower conductivities 2.74 to 4.3 mS/cm for raw milk samples were also reported in Morocco²⁰.



Mean monthly variation in Conductivity

Bacteriological quality of raw milk and milk products was assured by Standard plate count method The overall standard plate count of raw milk samples collected from vendors for two consecutive years shows minimum 5.13 and maximum 7.26 with a mean of 6.13 ± 0 . 083 log ₁₀ Cfu/ ml. In case of milk vending shops average standard plate count of raw milk samples for two consecutive years shows minimum 5.60 and maximum 7.50 with a mean of $6.37\pm0.085 \log_{10}$ Cfu/ ml table-1). Results

of Analysis of Variance test of the data revealed that there is highly significant (P<0.05) difference between the mean SPC count of samples belonging to different sources. Overall mean standard plate counts recorded in this study for raw milk from all sources were higher than the Indian standards (>10⁵) set by BIS for raw milk. Chandra Shekhar *et. al.*, reported these counts with a mean of $5.50\pm0.99 \log_{10}$ Cfu/ml from raw milk samples collected from vendors of Faizabad district of Uttar Pradesh²¹. The findings of the present study are in concurrence with their findings.

Coliform bacteria are associated with fecal and environmental contamination. Mean annual Coliform counts for the samples of raw milk collected from milk vendors were ranged between 3.20 to 4.23 with a mean of $3.86 \pm 0.1165 \log_{10}$ cfu/ml in the year 2009, where as it ranges from 3.46 to 4.97 with a mean of $4.16\pm0.1660 \log_{10}$ Cfu/ml in the year 2010 (table-1). The overall mean Coliform count of samples for both the year shows minimum 3.33 and maximum 4.65 with a mean of 4.01 ± 0.1413 log₁₀ Cfu/ ml. In the case of milk vending shops Coliform count ranged from 3.36 to 5.36 with a mean of $4.27\pm0.1761 \log_{10}$ Cfu/ml in the year 2009. Coliform counts for year 2010 varied from 3.55 to 5.42 with a mean of 4.44±0.1839 log₁₀ Cfu/ ml. Average Coliform count of milk samples for both the years shows maximum 5.39 and minimum 3.46 with a mean of 4.36± 0.180 log₁₀ Cfu/ ml. results of Statistical analysis of the Coliform counts also revealed significant (P<0.05) difference between counts of milk samples from both sources. Coliform counts obtained in this study were higher than reported earlier. This is an indicator of unsanitary conditions or practices during production processing and distribution or storage of raw milk. Inadequate cooling of milk and udder infections were also responsible for these higher counts.²⁴

Conclusion

The present study reveals that the quality of raw milk in Bhopal city was not satisfactory. It is confirmed by the high values of total bacterial count, Coliform counts and physico-chemical tests. Presence of such higher counts causes deterioration in the quality of raw milk. In conclusion, it can be said that the raw milk sold in the study area may pose a potential public health risk and therefore, hygienic precautions should be taken by determining critical control points from phases of production, storage and sale.

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Dacter lological counts of Taw mink samples during 2007-2010							
Bacteriological	Sauraa	Dunation	Ν	No. of colonies (log ₁₀ cfu/ml)			
Parameters	Source	Duration		Minimum	Maximum	Mean	±SE
Standard plate counts	Milk vendors	2009	60	4.97	7.15	6.14	0.085
		2010	60	5.28	7.36	6.11	0.081
	Average count		120	5.13	7.26	6.13*	0.083
	Milk vending shops	2009	60	5.56	7.72	6.34	0.095
		2010	60	5.63	7.26	6.39	0.075
	Average count		120	5.60	7.50	6.37*	0.085
Coliform counts	Milk vendors	2009	60	3.20	4.23	3.86	0.1165
		2010	60	3.46	4.97	4.16	0.1660
	Average count		120	3.33	4.65	4.01*	0.1413
	Milk vending shops	2009	60	3.36	5.36	4.27	0.1761
		2010	60	3.55	5.42	4.44	0.1839
	Average count		120	3.46	5.39	4.36 *	0.180

Table-1Bacteriological counts of raw milk samples during 2009-2010

N = Total no. of sample. Figure bearing * differ significantly at P< 0.05

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