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# BACTERIOLOGICAL QUALITY AND SAFETY OF RAW MILK IN KASHMIR VALLEY

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The microbiological safety of raw milk from twenty dairy farms in Kashmir valley was determined. Milk samples were collected from four districts namely Pulwama, Baramulla, Srinagar and Ganderbal of the Kashmir valley according to stratified random sampling design. Samples were analyzed for Coliforms, Standard Plate Count (SPC) and Methylene Blue Reductase Test (MBRT). It was observed that there is non-significant difference ($p>0.05$) in flavor, appearance and MPN index of milk samples of four districts. Further, SPC on $10^{-7}$, $10^{-6}$ and $10^{-5}$ dilutions differ significantly in all districts of the valley. Results from the Analysis of Variance (ANOVA) suggested that there was a significant difference ($p<0.05$) in the quality of milk among the four districts using various methods for acceptance and rejection of milk.

**Keywords**: Raw milk, Microbiological quality, Coliforms, Kashmir, Physiochemical properties

Milk, according to the PFA Rules, is the normal mammary secretion derived from the complete milking of healthy milch animal without either addition thereto or extraction therefrom. The health benefits of milk and dairy products are known to humanity since medieval times and may be attributed to the biological active components that are present in milk and also, due to their suitably modulated activities produced through the action of probiotics (Abdel-Salam 2010). Milk is a complex mixture of specific bioactive proteins, lipids and saccharides and contains numerous biologically active substances such as immunoglobulins, enzymes, antimicrobial peptides, Oligosaccharides, hormones, cytokines and growth factors (Pouliot and Gauthier 2006). Any deterioration in the quality of milk may affect the health of people consuming it including the vulnerable sections of the society: the growing child, the expectant mothers and senior citizen of the country. Milk is a highly nutritious food for human beings, but it also serves as a good medium for the growth of many micro-organisms. The detection of coliform bacteria and pathogens in milk indicate a possible contamination of bacteria either from the udder, utensils or water supply used. However, keeping milk in clean containers at refrigerated temperatures immediately after milking process may delay the increase of initial microbial load and prevent the multiplication of microorganism in milk between milking at the farm and the transportation to the processing plant **(**Adesiyun 1995). Contamination of

  

mastitis milk with fresh clean milk may be one of the reasons of the high microbial load of bulk milk (Jeffery and Wilson, 1987). The presence of pathogenic bacteria in milk emerged as major public health concerns. Especially for those individuals who still drink raw milk (Ryser, 1998). Most recently, E. coli 0157 has become a serious threat to the dairy industry with several outbreaks reported in developed countries ranging from mild diarrhea to potentially fatal hemolytic uremic syndrome (HUS), hemorrhagic colitis and thrombotic thrombocytopenic purpura (Coia et al., 2001). The enumeration of E. coli in foods is usually carried out by the conventional most probably number (MPN) procedure. At present some alternative methods based on direct plate on desoxycholate are also used for this purpose. Milk from a healthy udder contains very few numbers of bacteria ( < $3x10^4$cfu/ml) but may become contaminated by microorganisms from the surrounding environment during milking and milk handling, from water and milk equipments (Cousins and Bramley, (1981)). Dairy products quality defects have been attributed to poor microbiological quality of raw milk and heat resistant enzymes (Marshall 1982, Muir et al., 1986). The production of high quality milk should therefore be priority for good quality end products of long shelf life and for marketing of value added products. This is generally not easy to achieve in developing countries due to factors such as poor hygiene and sanitation during milking and milk handling, unclean water, high ambient temperatures, lack of cooling facilities and inadequate infrastructure for milk transportation to the processing facilities (Bille et al., 2009).

India is the second largest producer of milk (FAO 2010). As milk is consumed by people of all the age groups, the wholesomeness and quality of milk is of great importance. Quality here includes the nutritional composition, microbial quality, presence or absence of adulterant and extent of pesticide residues in milk. Kashmir Valley is not more deficit in the production of milk which is now established fully from the fact that during last year, the milk plant at Cheshmashahi, Srinagar Kashmir exported more than 1 lakh liters of raw milk to Mother Dairy, Delhi and the trend continues till date. The main objective of this study was to assess physical, chemical and microbial properties of raw milk of the Kashmir valley.

## MATERIAL AND METHODS

A total of 40 raw milk samples were collected from dairy farmers who use to send their milk to local market of Srinagar. Farmers involved in the study were chosen according to stratified experimental design whereby the Kashmir valley was divided into 4 specific regions, namely southern (Pulwama district) Northern (Baramulla district), Eastern (Ganderbal district) and central (Srinagar district) over a period of four months from November 2010 to March 2011. Approximately 100-300 ml milk was aseptically sampled from containers (buckets or churns) of bulk milk from each individual farmer into a sterile bottle in the early morning. It was collected immediately after milking, using hand into bulk milk container at ambient temperature. Samples were delivered to the laboratory in a cool box within 1-2 hour of collection and tested immediately. A total of 40 samples, ten samples from each district (Pulwama, Baramulla, Srinagar and Ganderbal) were taken for bacteriological analysis. The samples were examined for acceptance or rejection of milk using standard procedures like Rapid rejection tests, Platform tests, SPC, MBRT, Caliform tests using the methodology of APHA – AWWA-WEF (1998). The MBRT is a rapid way of determining quality of milk. The time taken by dehydrogenises produced by bacteria, to reduce methylene blue dye and decolonize it is an indication of degree of bacterial contamination.

The choice of method is influenced by such factors as the general quality of the

 

supply, cost, interest in quality improvement etc. Finally, the data obtained was carefully statistically analyzed through software package SPSS 16.0 version.

| MBRT | Classification | Approximate No. of Bacteria/ml |
|---|---|---|
| 0-30 minutes | Very poor quality | >20,000,000 |
| 31-120 minutes | Poor quality | >4,000,000 |
| 121-360 minutes | Fair quality | >500,000 |
| 361-480 minutes | Good quality | <500,000 |

*Source: pp-206 Food Microbiology and Safety Practical Manual 2005.*

## RESULTS AND DISCUSSION

The MPN technique consists of an estimation of the density of viable organisms in a sample and is particularly useful for low level of micro organisms. It provides as an index of hygienic standard used in the production of milk, as unclean udder and teats can contribute the presence of coliform from various sources such as manure, soil, feed, personnel and water. Out of the five raw milk samples taken from each district, the corresponding mean counts of the milk from the 4 districts i.e., Pulwama, Baramulla, Srinagar and Ganderbal are as 115.11 $\pm$ 7.09, 141.11$\pm$14.06, 165.50$\pm$10.08, and 81.30 $\pm$ 12.59 respectively. The highest MPN index was of Srinagar and lowest was that of Ganderbal as shown in Table 1 ($p<0.05$). Coliforms are widely used indicators of faecal contamination as they make up about 10% of the intestinal microflora of human and other animals. Their enumeration is usually carried out by multiple tube fermentation tests. *E. coli* is frequently used in the microbiological analysis in any food as an indicators organism of poor hygienic conditions.

It is observed from Table 2 that out of the four districts, the standard plate count when inoculation was done with $10^{-7}$ dilution, district Ganderbal has the highest mean count (346.61 $\pm$ 6.61) per ml and lowest mean count was found in district Pulwama (284.41 $\pm$ 4.07).

Table 1. Mean $\pm$ S.D (n = 10) of MPN counts of raw milk obtained from 4 districts

| Districts | Pulwama | Baramulla | Srinagar | Ganderbal |
|---|---|---|---|---|
| **Mean $\pm$ S.D** | 115.11 $\pm$ 7.09 | 141.11 $\pm$14.06 | 165.50 $\pm$10.08 | 81.30 $\pm$ 12.59 |

In case of $10^{-6}$ dilution the highest mean bacterial count was observed in district Ganderbal (449.51 $\pm$ 20.13) per ml and lowest was obtained from district Baramulla (379.41 $\pm$ 12.77) and in case of dilution $10^{-5}$ district Srinagar has the highest bacterial count (553.02 $\pm$ 20.53) per ml and Ganderbal has the lowest (459.91 $\pm$ 14.01). Statistically significant difference was observed in all districts when inoculation was done with different dilutions ($p<0.05$).

Table 2. Mean $\pm$ S.D (n = 10) of SPC of milk samples

| $10^{-7}$ dilution | | | | |
|---|---|---|---|---|
| Districts | Pulwama | Baramulla | Srinagar | Ganderbal |
| **Mean $\pm$ S.D** | 284.41 $\pm$ 4.07 | 327.52 $\pm$ 6.11 | 337.01$\pm$ 4.96 | 346.61 $\pm$ 6.61 |
| $10^{-6}$ dilution | | | | |
| Districts | Pulwama | Baramulla | Srinagar | Ganderbal |
| **Mean $\pm$ S.D** | 411.61$\pm$ 19.29 | 379.41 $\pm$ 12.77 | 434.42 $\pm$ 15.07 | 449.51 $\pm$ 20.13 |
| $10^{-5}$ dilution | | | | |
| Districts | Pulwama | Baramulla | Srinagar | Ganderbal |
| **Mean $\pm$ S.D** | 499.01 $\pm$ 19.06 | 530.01 $\pm$ 15.12 | 553.02 $\pm$ 20.53 | 459.91 $\pm$ 14.01 |

The reason for the high counts could be due to infected udders of the cows,

unhygienic milking procedures or equipments, and or inferior microbiological quality of water used for cleaning, utensils and animals as well as the milk storage conditions. Therefore poor milk quality has been considered as one of the major reason for losses and results in deduced income for the small farmers in Kashmir.

The milking process, especially the equipment associated with it, introduces the greatest proportion of microorganisms in raw milk (Cousin, 1982). The use of detergents and good quality water for cleaning the equipment could be expected to remove milk remains including microorganism and thereby affects the microbiological quality of milk. High numbers of bacteria may be associated with unclean udder, teats and unclean storage tanks (IDF). Practicing very good hygienic principles at the farms, in handling and transportation of milk, is a must. It is important to cool and store raw milk quickly. Equipments that is poorly designed with respect to cleaning and potential for fouling represent another hidden source of psychotropic and thermophilic bacteria, as well as the destructive enzymes.

Table 3. Mean ± S.D (n = 10) of MBRT of raw milk samples

| Districts | Pulwama | Baramulla | Srinagar | Ganderbal |
|---|---|---|---|---|
| **Mean ± S.D** | 315.25 ± 13.01 | 301.02 ± 11.33 | 278.51 ± 13.83 | 241.12 ± 12.45 |

Table 3 reveals that the highest mean reduction time is observed in Pulwama district as 315.25 ± 13.01 and lowest MBRT was found in samples pertaining to Ganderbal with a mean of 241.12 ± 12.45. The Analysis of Variance (ANOVA) suggested that there was a significant difference ($p<0.05$) in the quality of milk in different districts using MBRT. The results obtained in this paper are in conformation with the results obtained by Chye et al., (2003), Saxena, and Agarwal (2004), ChatterjJee et al., (2006) and Bille et al., (2009).

## CONCLUSION

Foods are not intended to only satisfy hunger and to provide necessary nutrients for humans now a day but also to prevent nutrition-related diseases and improve physical and mental well-being of the consumers (Menrad, 2003). It is now established fully that Kashmir valley is not more deficit in the production of milk as per the reports from Animal Husbandry department. Consumer demands in the field of food production have changed considerably in the last two decades. Consumers more and more believe that foods contribute directly to their heath (Elsanhoty et al., 2009). In the present investigation, it was concluded that all districts seem to supply milk below the standard set by the Department of Veterinary Services. To prevent flavor/odor defects in milk, proper milk handling procedures from the farm to the consumer are essential. As quality improvement progresses, interest goes far beyond rapid rejection tests. The highest bacterial load was found in the samples taken from Ganderbal and lowest load in the samples taken from Pulwama district on the basis of MBRT; as it is the platform test used for determining the quality of milk. The SPC was highest in samples taken from Ganderbal on $10^{-7}$ dilution, and lowest in Pulwama; the SPC on $10^{-6}$ dilution was highest in samples taken from Ganderbal and lowest in Baramulla also the SPC on $10^{-5}$ dilution was highest in samples taken from Srinagar district and lowest in Ganderbal. Animals suffering from certain diseases may transmit them through their milk to man, although this method of infection is less common than that through milk to which germs have had access in process of handling or transportation. The principal diseases which may be derived from the cow through her milk are tuberculosis and diphtheria. Regular sterilization of dairy equipment, washing of utensils, milkers hand, udders, eradication of diseased animals,

 

pasteurization/ boiling of milk is required before collection and distribution for consumption and product making. The best way to ensure safety of milk and good health is by strict control and maintenance of hygiene at all stages of processing and distribution coupled with improved personnel hygiene. The present study recommended that quality assurance programs should ensure good quality milk and milk products.

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