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MICROBIAL QUALITY OF RAW COW MILK SAMPLE COLLECTED FROM UPPARAPALAYAM AND AARIKKAMEDU VILLAGES OF THIRUVALLUR DISTRICT, TAMILNADU, SOUTH INDIA

Kavitha*

Assistant Professor, PG and Research Department of Zoology, Quaid-E-Millath Government College for Women (Autonomous), Chennai- 600 002.

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***Corresponding Author**
Kavitha
Assistant Professor, PG and Research Department of Zoology, Quaid-E-Millath Government College for Women (Autonomous), Chennai- 600 002.

ABSTRACT

Milk of cows has long been considered a highly nutritious and valuable human food, and is consumed by million daily in a variety of different products. Milk being nutritious food for human beings, also serves as a good medium for the growth of many microorganisms. Raw milk, as it leaves the udders of healthy animals normally contains very low numbers of microorganism. This study was aimed at determining the microbial quality of raw milk of cows collected from Upparapalayam and Aarikkamedu villages of Thiruvallur District, TamilNadu. Dairy cattle raised under different systems of rearing were identified and classified into three group's viz., Housed under the sky, Housed in mud flooring shed, Housed in concrete flooring shed. The indigenous dairy cows, cross bred cows and buffaloes were grouped separately and thirty three animals represented for each group. Methylene blue reduction test (MBRT) and microbiological quality of each sample was analyzed using standard procedures. The MBRT values, Standard plate count and total coliforms were significantly above the normal recommended levels were observed in the current study. This may be due to poor milk handling practices during milking, poor animal health services, and use of poor potable water. High microbial counts and the occurrence of pathogens are likely to affect the keeping quality and safety of raw milk as well as products derived from it. The achievement of hygiene in dairy farm directly influences the production oriented economic results and health safety perspectives in human beings. It is therefore critically important to ensure high quality raw milk production from healthy animals under good hygienic conditions and to apply control measures to protect human health.

KEYWORDS: Raw milk, Methylene blue reduction test, Coliforms, Standard plate count.

INTRODUCTION

Milk is a compulsory part of daily diet for the expectant mothers as well as growing children.[1] According to Ramesh,[2] the major components of milk are water (87.4%), milk solids (12.60%), solids-not-fat (9.0%), fat (3.60%), protein (3.40%), milk sugar or lactose (4.90%) and ash or minerals (0.70%). Tamilnadu, the southernmost state of India is one of the top ten milk producing states in the country with an annual milk production of 4.75 million tones.[3] Milk is a complex biological fluid and by its nature, a good growth medium for many microorganisms. Because of the specific production it is impossible to avoid contamination of milk with micro-organisms.[4] Bacterial contamination of raw milk can originate from different sources such as air, milking equipment, feed, soil, faeces and grass.[5,6] Raw milk carries harmful bacteria causes' diarrhea, stomach cramping, and vomiting, rarely kidney failure, paralysis, chronic disorders, and even death. In the present study microbiological quality of milk produced under various production systems at village and town levels were assessed. All cases of dairy illness continued to be of bacterial origin, pathogens that have involved in communicable diseases associated with the consumption of milk include *Salmonella*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Campylobacter*, *Yersinia*, pathogenic *Escherichia coli* and *Clostridium botulinum*.[7,8] The detection of coliform bacteria, pathogens and high microbial count in milk are major factors in determining the quality of milk. It indicates the hygienic level exercised during milking, that is, cleanliness of the milking utensils, condition of storage, manner of transport as well as the cleanliness of the udder of the individual animal. Milk from a healthy udder contains few bacteria but it picks up many bacteria from the time it leaves the teat of the cow until it is used for further processing. Coliforms are indicators of both the manner of handling milk from milking till consumption and the quality of the milk. Milk produced under hygienic conditions from healthy animals should not contain more than 5x105 cfu/m.[9] Hence the present study was undertaken to elucidate the microbiological quality of raw cow milk reared near Upparapalayam and Aarikkamedu villages of Thiruvalloor District.

2. MATERIALS AND METHODS

Description of the study area

The study was conducted in Upparapalayam and Aarikkamedu villages of Thiruvallur District, TamilNadu. Tiruvallur district is an administrative district in the South Indian state

of Tamil Nadu. The town of Tiruvallur is the district headquarters. The district has a mixture of urban and rural characteristics. The Eastern part of Tiruvallur district is dominated by urban characteristics while the Southern and Northern part of the district has influence of Andhra culture due to its position.

Experimental Design

Dairy cattle raised under different systems of rearing were identified and classified into three groups viz., Housed under the sky, Housed in mud flooring shed, Housed in concrete flooring shed. The indigenous dairy cows, cross bred cows and buffaloes were grouped separately and thirty three animals represented for each group.

Data collection and milk sampling

Fifteen ml of milk was collected from each dairy cattle. The samples were collected as per the recommendations of Bureau of Indian Standards, IS: 1479 (Part I)-1960 (Methods for Dairy industry; Part I. Rapid examination of milk)

Methylene Blue Reduction Test (MBRT)

Details of the apparatus reagents and general procedure were same as described in 12 of IS: 1479 (part I)-1960. The tubes were incubated beyond half an hour until the milk was completely decolourized or decolourized up to 5 mm from the surface. The tubes were observed and inverted initially at half hour and at hourly intervals thereafter. If the milk begins to decolourize, the tubes should not be inverted. Recorded the time at which complete decolourization was observed. The dye reduction time refers to the microbial load in the milk and the total metabolic reactions of the microorganism.[10]

Grading

The following methylene blue reduction times were suggested as guide for grading.

Decolorization time in Hours	Milk Grade
5 and above	Very good
3 and 4	Good
1 and 2	Fair
½	Poor

Standard Plate Count

This method was used for determining the total number of viable bacteria in milk and consist of mixing appropriate quantity of milk with suitable nutrient agar medium in a Petri dish and counting the bacterial colonies developed after incubation at specified temperature for a specified temperature for definite period of time. The plate count of bacteria in milk was determined according to the IS 5102 -1969.

Preparing Samples for Plating

Immediately before removal of any portion of any sample, vigorously mix the contents of each container till thorough mixing is assured. Invert the filled retail containers until contents are homogeneous before removal of test portions. Once opening a sample from the container, remove from closure all material which may contaminate the sample. Where necessary, wipe the top of opened container, with sterile cloth saturated with alcohol (BIS:1479). Presence of bacterial count less than 5 millions is taken as satisfactory.

Coliform Bacteria

This group of bacteria is important in quality control of milk as it is indicative of possible faecal contamination and due to the ability to produce acid and taints in milk. The presence of this group milk considered to be an indicator of the degree of unhygienic practices during production, processing or storage and is intended to measure general care taken in handling this product. The members of this group are generally destroyed during pasteurization treatments and, therefore positive coliform test in the case of pasteurized milk is considered indicate post-pasteurization contamination. The coliform bacteria in milk was determined in accordance with the procedure laid down in IS: 5401-1969.

Interpretation

The following coliform contents are suggested as a guide grading of raw milk supplies:
Raw Milk:'Coliforms' < 1000 cells /ml – satisfactory.

In case of milk containing added sugar, the results of the presumptive coliform test shall be interpreted with caution because of the suitable risk of a false positive.

Statistical analysis

The data were analyzed statistically, using ANOVA and the results were discussed.

RESULTS AND DISCUSSION

Methylene Blue Reduction Test (MB)

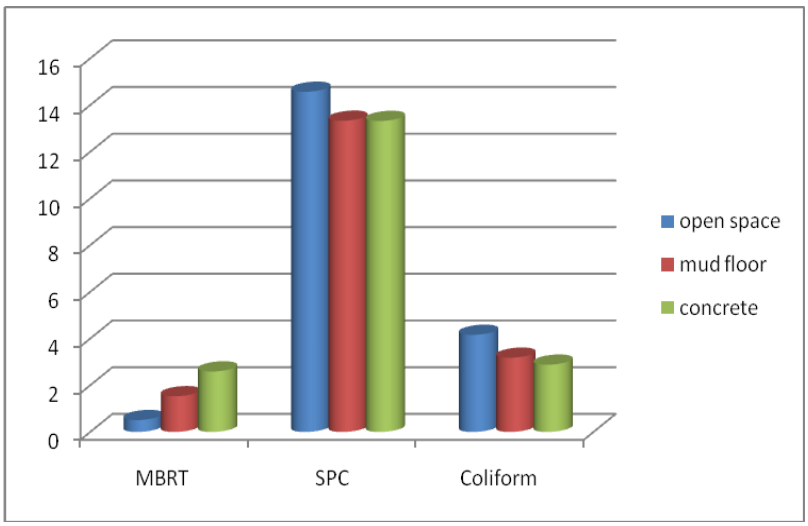
Methylene Blue dye has been employed to check for the overall microbial load and quality control of milk and other liquid foods.[11] The MBRT values are significantly above the normal recommended levels were observed in the current study. MBRT for indigenous cows under the sky is 0.51 ± 0.04, under mud flooring shed being 1.53 and under concrete floor shed 2.59 ± 0.14 hrs. Regarding crossbred animals, the values are 1.58± 0.10, 2.21± 0.021 and 3.11± 0.22 hrs respectively. For buffalos the values are 2.45± 1.22, 2.58± 0.16 and 3.58± 0.17 hrs. All the values are significantly above the normal recommended level of bacterial load for raw milk as per Bureau of Indian Standards. The values also differ significantly among the groups. These finding correlate with the values of previous study by Muhammad *et al*.[12] Bongard *et al*.[13] Merker *et al*.[14] reported that blue color disappearance in short time indicates higher microbial load in the milk sample In this study, most of cow's raw milk shows very short decolorization time of the dye. This may be due to poor milk handling practices during milking, poor animal health services, and use of poor potable water.

Standard Plate Count (SPC)

Standard Plate Count is the most frequently used in regulatory programs and reflects the general hygienic condition during milk production, while each of the other tests identify a potential contamination source of concern for milk quality.[15] The standard plate count method is used for determining total number of viable bacteria in milk. In the present study, the standard plate count for indigenous cattle housed under the sky, mud flooring shed and concrete flooring are 14.57± 0.12, 13.33 ± 0.27 and 13.32± 0.27 (1×106/ml) observed respectively. The values of bred cows are 13.17± 0.34, 12.91± 0.31 and 12.4 ± 0.31 (1×106/ml) respectively. Whereas for buffaloes, the values are 12.70 ± 0.39, 12.47 ± 0.23 and 10.29 ± 0.45 (1×106/ml). All the values are significantly higher than the specified value of 5 millions (for poor quality).The values also differ significantly within the groups without any particular pattern of interpretation with respective the housing environment of the cows. This means the microbial contamination would be from multifarious sources like environmental load due to poor ventilation, dusty environment and poor external udder hygiene. These findings agrees with result of the previous study by Kashifa *et al*, Zweifel *et al*. and Lekh *et al*.[16,17,18]

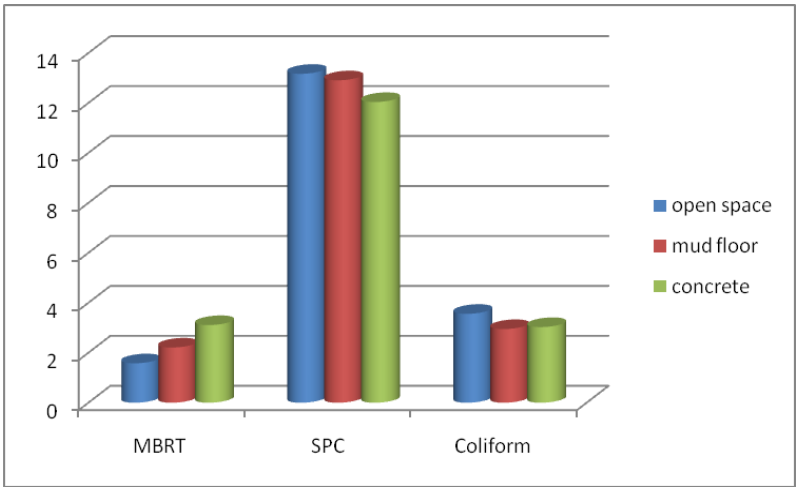
Coliform Count (CC)

Coliforms are indicator of poor hygienic. The increased number would commonly be due to contamination of fecal and bedding materials. In this study the coliform count of raw of indigenous animals housed under the sky, mud flooring and concrete flooring are 4.16 ± 0.41, 3.18 ± 0.07 and 2.88 ± 0.15 (1×103/ml) respectively. For cross bred cows the values of coliform in raw milk are 3.56 ± 0.23, 2.95 ± 0.12 and 3.03 ± 0.13 (1×103/ml) respectively and for buffalos the values are 2.32 ± 0.32, 2.49 ± 0.16 and 2.07 ± 0.17 (1×103/ml) respectively. All the values are significantly higher than the acceptable count of 1×103 per ml. The coliform count of cows housed on concrete flooring shows lesser values when compared to the other system of housing, but only insignificantly. This shows the udder health, milking hygiene, fecal contamination from the bedding materials remains inevitable inspite of different system of housing. The results of this study fall in line with the previous studies by several researchers,[19,20,21] The source of contamination is well documented by Hogan *et al*.[22] Olson and Mocquot opined that the detection of coliform bacteria and pathogens in milk is an indication of possible contamination of bacteria either from the udder, milk utensils or water supply used.[23] The coliform count obtained in the current study is greater than that reported by Fekadu[24] who found coliform counts from cows' milk produced in Aneno, Gulgula and Dongora districts of southern region, respectivel The higher coliform count observed in this study may be due to the initial contamination of the milk samples either from the cows, the milkers, milk containers and the milking environment.



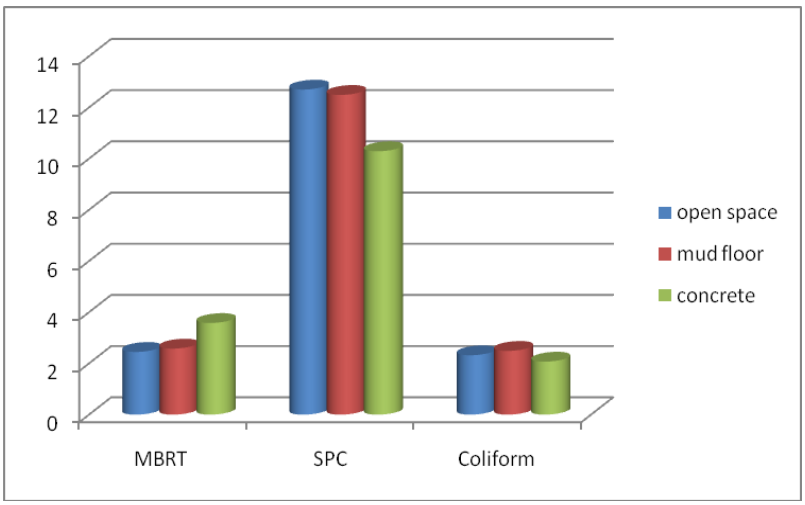
MBRT in Hours SPC in 10^6/ml Coliform in 10^3/ml

Fig. 1: Microbial Quality of Indigenous cows' milk.



MBRT in Hours SPC in 10^6/ml Coliform in 10^3/ml

Fig. 2: Microbial Quality of Cross bred cows' milk.



MBRT in Hours SPC in 10^6/ml Coliform in 10^3/ml

Fig. 3: Microbial Quality of Buffaloes' milk.

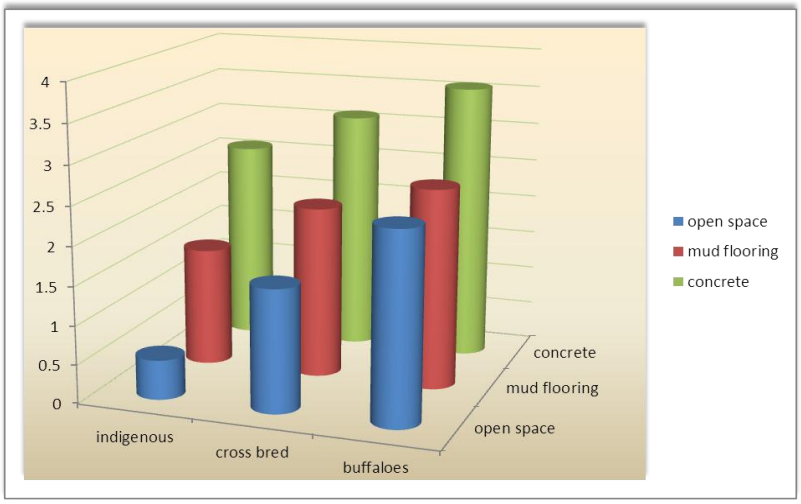


Fig. 4: Methylene Blue Reduction Time (in Hrs) of raw milk under different housing systems.

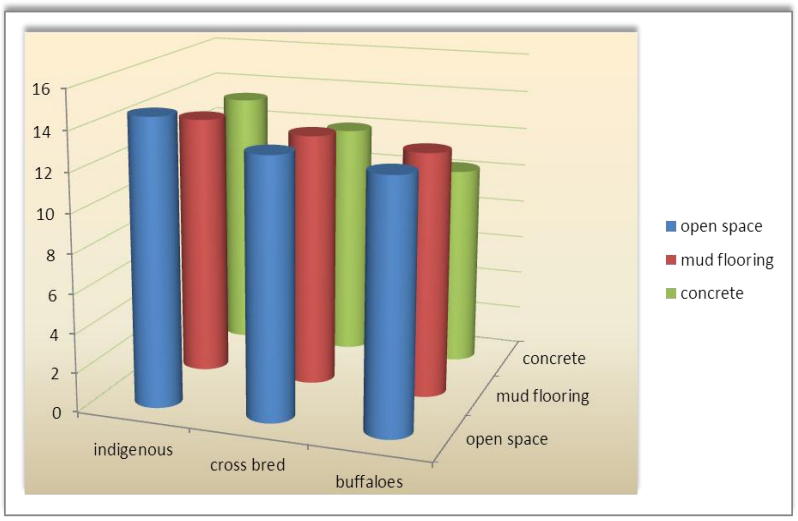


Fig. 5: Standard Plate Count of raw milk (1×10^6) under different housing systems.

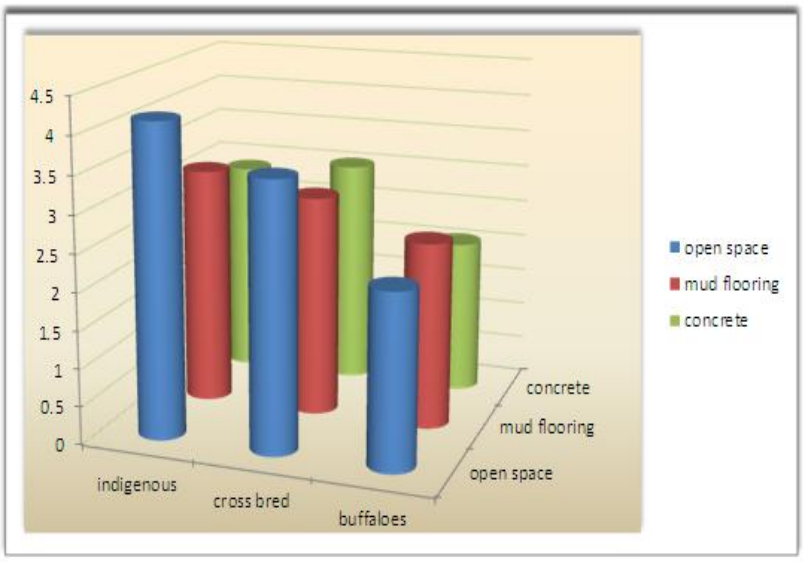


Fig. 6: Coliform count of raw milk (1×10^3) under different housing systems.

CONCLUSION

The present study showed that the quality of milk produced in the study area was poor. This was evident from the high values of Methyline blue reduction test, standard plate count and coliform could be attributed to poor management, unhygienic environment of shed, lack of awareness of preparing the udder before milking, faulty milking techniques, poor hygiene of the hind quarter and improper cleaning utensils. The poor bacteriological quality observed in the present study requires further investigation of the status of the animals' health, especially mastitis and the significance of the effect of containers to ascertain their contribution on microbial quality. More attention should be focused on the cleaning of transportation tanks, appropriate handling with milk and its transportation at low temperatures from farms and

collecting points to the dairies, particularly in summer season. Therefore, it is recommended that training and guidance should be given to farms owners and their workers responsible for milking. Meanwhile, information on health hazards associated with contaminated raw milk should be extended to the public, so that consumption of untreated/improperly treated raw milk could be avoided.

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