



Original Research Article

## A Cross sectional Study to assess Quality of Informally Marketed Milk in Urban & Rural Field Practice Areas of J.N.M.C., Belgaum

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### ABSTRACT

**Background:** Milk being very nutritional and balanced foodstuff, is a well-known medium that favours the growth of several microorganisms. Many milk borne epidemics of human diseases spread by contamination of milk by spoiled hands of dairy workers, unsanitary utensils, flies and polluted water supplies.

**Objectives:** The study was designed to assess the quality of informally marketed raw milk in rural and urban households.

**Methodology:** A cross sectional study was conducted among 500 households, 250 (20% of all households) each residing in urban and rural areas, Belgaum district using predesigned and pretested questionnaire. Informally marketed raw milk samples were collected from 10% households (25 houses) each from both the areas for laboratory analysis using Brucella milk ring test (BMRT), Methylene blue reduction test (MBRT) and Coli form tests (CT) at the teaching hospital.

**Results:** None of the participants knew that raw milk can transmit diseases, however 28% of urban and 38% of rural participants knew that milk could be contaminated. 32% urban and 62.4% rural participants reported to consume raw milk, primary reason being healthy and convenient. 13.6% of urban and 47.2% of rural households possessed their own milk producing animals.

Laboratory analysis of 25 urban & 25 rural milk samples showed, mean specific gravity of  $1.026 \pm 0.004$  and  $1.025 \pm 0.007$  respectively. 5 (10%) samples were positive for BMRT and 14 (28%) were positive for MBRT as well as CT. Samples were considered positive for CT when viable count was  $>10^5$  bacteria/ml of milk. Urban and rural buffalo milk samples showed significant association with the MBRT and CT.

**Conclusion:** Awareness of milk borne diseases was nil among the study participants and milk contamination was found more in rural area.

**KEYWORDS:** Informally marketed milk, Brucella milk ring test, coli form test, methylene blue reduction test

## INTRODUCTION

Milk being nutritional and balanced foodstuff, is a well-known medium that favours the growth of several microorganisms. <sup>[1]</sup> Many milk borne epidemics of human diseases spread by contamination of milk by spoiled hand of dairy workers, unsanitary utensils, flies and polluted water supplies. <sup>[2]</sup>

India's milk production has grown at around 4% annually in the recent years, which far exceeds the global average of about 1%. <sup>[3]</sup> India keeps over three times the number of cattle as the USA. In addition, 94 million buffaloes contribute to milk production in India. It is estimated that around 15% of the milk produced in India is marketed through formal channels, while the remaining 85% is informally handled. <sup>[4]</sup>

The informal milk market pathways persist because they provide social and economic benefits to smallholder producers, small market agents and consumers in terms of higher farm gate prices, creation of employment and competitive consumer prices. <sup>[5]</sup>

Risks of milk-borne zoonoses posed by the informal market are amplified by poor handling procedures in the market, the lack of quality standards and the fact that most consumers prefer raw milk over pasteurized milk. <sup>[1]</sup>

There is limited data existing on raw milk consumption and corresponding risks of milk borne illnesses. It is a highly perishable commodity and poor handling can exert both a public health and economic toll, thus requiring hygienic vigilance throughout the production to consumer chain. <sup>[5]</sup>

As classified by the Joint FAO / WHO Expert Committee (1970) on milk hygiene; the most important milk borne diseases transmitted from animals to humans are: brucellosis, tuberculosis, streptococcal infection, staphylococcal, salmonellosis and

Q fever. Diseases of less importance are - cow pox, foot and mouth disease, anthrax, leptospirosis and tick borne encephalitis. Infections transmitted through milk are - typhoid, paratyphoid fevers, shigellosis, cholera and entero-pathogenic E coli. <sup>[6]</sup>

The purpose of this study was to assess the milk quality regarding adulteration with water, contamination with E. coli and brucella.

## METHODOLOGY

The present study was conducted in rural and urban field practice areas of Dept of community medicine, Jawaharlal Nehru Medical College (J.N.M.C); Belgaum district, Karnataka, South India. A total of 20 % each (250 households) of total households were selected by systematic random sampling to collect data by predesigned and pretested questionnaire from rural and urban areas. Of these 10 % of households, i.e. 25 houses each from both the areas, using informally marketed milk were selected by simple random sampling and raw milk samples were collected for laboratory analysis. Written informed consent was obtained. Institutional Ethics Committee clearance was taken from J.N.M.C Belgaum, Karnataka.

The information thus collected was computerized and analyzed by using Statistical Package for Social Science (SPSS 10.0) software program for Windows. Data was expressed in terms of rates, ratios and percentages. Laboratory reports were analyzed separately. Statistical analysis was done using Chi Square test and Fisher exact test. A probability value (p value) of less than 0.05 was considered as significant.

### ***Milk samples were subjected to following tests***

Physical test:

- Specific gravity of milk by lactometer

This was measured at the point of collection by the investigator using lactometer. Specific gravity of milk was measured using lactometer of Amber Company to detect the change in density of adulterated milk with water. Milk sample was gently poured into a measuring cylinder (50 mL). The lactometer was left to sink slowly into the milk. Measurement was read and recorded to the last Lactometer degree ( $^{\circ}$ L) (30) just above the surface of the milk. For the calculations, lactometer degrees were used, and for the conversion to density 1.0 was written in front of the true lactometer reading, that is, 1.030 g/mL. The average specific gravities considered were;

- Cow Milk - 1.028 to 1.030
- Buffalo Milk - 1.030 to 1.032
- Goat Milk - 1.028 to 1.030 <sup>[7]</sup>

#### **Microbiological tests:** <sup>[8]</sup>

##### ❖ *Methylene blue reduction test to test presence of bacteria*

10ml of milk and 1ml of methylene blue solution was added to the 20ml of sterilized test tubes. Then tubes were closed with sterile rubber stopper, slowly tubes were inverted once or twice and then kept in water bath. Test was considered positive when whole column of milk was decolorized within 30minutes.

##### ❖ *Coli form test to detect faecal contamination of milk*

Varying amounts of milk were added to tubes of bile salt lactose medium. For unknown quality of milk the following series was suggested (1 ml of milk in 9 ml of MacConkey broth)

- 1.0 ml of a 1 in 10 dilution of milk
- 1.0 ml of a 1 in 100 dilution of milk
- 1.0 ml of a 1 in 1,000 dilution of milk
- 1.0 ml of a 1 in 10,000 dilution of milk

The smallest amount that yields acid and gas was ascertained. Under the Scottish

regulations, for standard milk, these tubes were inoculated each with 1 ml of 1 in 1000 dilution. The milk sample was taken to have passed the test if acid and gas were absent from two of the three tubes. Samples were considered positive for coli form test, if showed more than  $10^5$  bacteria per ml of milk.

##### ❖ *Brucella milk ring test*

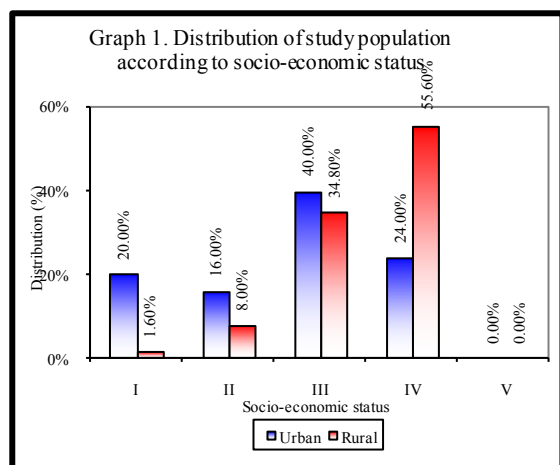
The milk was mixed thoroughly and poured into a test tube sufficient to give a column of milk about 1 in high. One drop of stained antigen was added and mixed thoroughly by shaking. Frothing was avoided which could interfere with reading of the test. It was incubated at  $37^{\circ}$  C water bath for about 40 to 50 minutes, which was sufficient time of the cream to rise.

In milk containing brucella agglutinins the bacteria were agglutinated and raised with the cream forming a blue cream line, having the skin milk white in samples, in which there were no agglutinins. There was a white cream line and the rest of the milk remained blue.

The results were interpreted as positive (+++). Cream layer formed a deep blue ring on top of a completely white column of milk. This indicated a high concentration of agglutinins. The white cream layer and milk column blue were considered as negative.

## **RESULTS**

Among all the households (500), Majority of study participants, 99.2% and 98.4% in urban and rural area respectively, responsible for handling of milk were females, 55% of urban and 77.2% of rural participants were illiterates. 44% households in urban area belonged to class III socio-economic status; where as 55.6% households in rural area belonged to class IV socio-economic status (Graph 1).



Many (32% urban and 62.4% rural) study participants consumed raw milk, primary reason being more healthy and convenience (12% in urban), health purpose (12% in rural) (Table 1). 32% of urban and majority rural participants had practice of adding water to milk (Table 2). Present study revealed 13.6% of urban and 47.2% of rural participants possessed their own milk producing animals.

None of the participants knew that raw milk can transmit diseases, however 28% of urban and 38% of rural participants knew that milk could be contaminated; of which 8% urban and 12% rural participants thought that milk utensil as main source of contamination. None of the participants suffered from milk borne diseases in the year preceding the survey.

Laboratory analysis of 25 urban & 25 rural milk samples showed, mean specific gravity were  $1.026 \pm 0.04$  and  $1.25 \pm 0.007$  in urban and rural area respectively. 10% samples (5 / 50) were positive for brucella milk ring test and 28% (14 / 50) samples were positive for both methylene blue reduction test and coli form test. Buffalo milk showed significant association with methylene blue reduction test and coli form test in both urban & rural areas (Table 3).

**Table 1: Distribution of study participants according to consumption of raw milk**

Consumption of raw milk	Reasons	Urban (n=250)		Rural (n=250)	
		Number	Percent	Number	Percent
Yes	Good taste	10	4.00	16	6.40
	Health purpose	10	4.00	30	12.00
	Convenience	10	4.00	18	7.20
	Taste and Convenience	20	8.00	15	6.00
	Health & convenience	30	12.00	15	6.00
<b>Total</b>		<b>80</b>	<b>32.00</b>	<b>156</b>	<b>62.40</b>
No		170	68.00	94	37.60
<b>Total</b>		<b>250</b>	<b>100</b>	<b>250</b>	<b>100</b>

**Table 2: Distribution of study participants according to knowledge regarding contamination of milk**

Contamination	Sources	Urban (n=250)		Rural (n=250)	
		Number	Percent	Number	Percent
Yes	Added water	10	4.00	16	6.40
	Utensils	20	8.00	30	12.00
	Dairy animal	0	0	1	0.40
	Human handling	20	8.00	17	6.80
	Water and dairy animal	10	4.00	15	6.00
	Added water and utensil	10	4.00	16	6.40
	<b>Total</b>		<b>70</b>	<b>28.00</b>	<b>95</b>

## DISCUSSION

The importance of milk in human diet is well established, as it is considered as the best, ideal and complete food for all age groups. However, in spite of being so, milk can also serve as a potential vehicle for transmission of some diseases under certain circumstances.

A study done in Ghana in 2003, reported that 46% respondents were males and 54% were females and in Tanzania 73% respondents were males compared to 27% females.<sup>[9]</sup> Present study showed majority participants handling milk were females, it may be because of working men.

Similar to finding of study done in USA in 2006, 42.3% of dairy produces

surveyed reported the taste and convenience as primary reason for consumption of raw milk. [10] Very similar findings found in various studies reported that 20% to 83% of

milk samples were adulterated with water, [1, 5, 9, 11,12] it might be because of lack of awareness about the milk borne diseases through contamination by water.

**Table 3: Association of laboratory tests with different types of milk samples.**

Laboratory tests	Result	Cow and goat's milk		Buffalo's milk	
		Urban (n=7)	Rural (n=12)	Urban (n=18)	Rural (n=13)
**Specific gravity	Normal	0 (0%)	8 (66.7%)	5 (27.7%)	1 (7.7%)
	Diluted	7 (100%)	4 (33.3%)	13 (72.3%)	12 (92.3%)
Brucella milk ring test	Positive	0 (0%)	1 (8.4%)	3 (16.7%)	1 (7.7%)
	Negative	7 (100%)	11 (91.6%)	15 (83.3%)	12 (92.3%)
Methylene blue reduction test	Positive	3 (42.8%)	2 (16.7%)	2 (11.2%)	7 (53.8%)
	Negative	4 (57.2%)	10 (83.3%)	16 (88.8%)*	6 (46.2%)
Coliform test	Positive	3 (42.8%)	2 (16.7%)	2 (11.2%)	7 (53.8%)
	Negative	4 (57.2%)	10 (83.3%)	16 (88.8%)*	6 (46.2%)

\* p<0.05 statistically significant using fisher exact test

\*\* Specific gravity – normal is non-adulterated and diluted is adulterated with water

Adulterated cow and goat's milk – < 1.028; non-adulterated milk – ≥ 1.028<sup>8</sup>

Adulterated buffalo's milk – < 1.030; non-adulterated milk – ≥ 1.030

A study done in Ghana and Tanzania in 2003 showed, 68% and 14% of the households had milk sourced from own animals. [9] Present study also revealed that 47.2% of rural and 13.6% of urban households possessed their own animals. This shows rural people prefer to have their own animals.

Various studies showed that 23% to 68.5% of the study participants were aware of diseases transmitted from milk. [10,13] In the present study there was lack of awareness regarding this. There is need of conducting health education camps in these study areas.

Various studies showed specific gravity of milk samples ranged from 1.027 to 1.030, [5, 9] while in the present study mean specific gravity were  $1.026 \pm 0.04$  and  $1.25 \pm 0.007$  in urban and rural area respectively. The specific gravity in the present study is low, might be because of their practice of addition of water to milk. Various studies reported 37% to 56% samples were positive for Brucella milk ring test. [5, 9] Present study showed 10 % of samples positive for Brucella milk ring test; it may be because sample size tested for laboratory tests was very less.

Various studies reported 20% to 100% samples were positive for E coli bacteria by coli form test. [14-18] Present study showed 28% (14 / 50) samples were positive for both methylene blue reduction test and coli form test. It might due to their practice of addition of contaminated water to milk. Laboratory analysis was limited to only 50 milk samples, due to operational constraints. And since we tested for pooled milk samples, it may not reflect the status of individual milk producing animal.

## CONCLUSION

This community based cross sectional study among 500 (urban and rural) households investigated the levels of knowledge, attitudes and practices regarding milk borne diseases. It revealed that none of them had knowledge about milk borne diseases. Many study participants consumed raw milk, primary reason being more healthy and convenience (12% in urban), healthy purpose (12% in rural). There is urgent need to conduct health education campaigns through Government or Private sectors, so that in future this contamination of milk can be reduced.

Because of less education and poor socio economic status contamination of milk was contaminated both in rural as well as urban areas. Laboratory analysis of 50 informally marketed raw milk samples revealed 5 (10%) were positive for brucella milk ring test and 14 (28%) were positive for both methylene blue reduction test and coliform test.

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*Conflicts of interest: none*

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