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RESEARCH ARTICLE

MICROBIOLOGYALGAE

MICROBIAL QUALITY OF RAW AND PASTEURIZED MILK SAMPLES COLLECTED FROM DIFFERENT PLACES OF WARANGAL DISTRICT, (A.P.) INDIA

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ABSTRACT

Milk is nutritious and essential food for human beings and also serves as good medium for microbial growth and contamination 240 raw milk samples and 72 pasteurized milk samples from different places of Warangal District for a period of six months were analysed for microbial quality. Among the raw milk samples only 19.1% of samples were good quality and 28.3% are very poor quality. In the pasteurized milk samples 81.9% of samples were good for human consumption. The bacteria isolated from milk samples includes *Lactobacilli, Staphylococcus aureus, Escheritia coli, Bacillus subtilis , Salmonella typhi,* and *feacal coliforms.*

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KEY WORDS

Raw milk, pasteurized milk, MBRT, coli forms, public health, Warangal.

INTRODUCTION

Milk is an essential part of daily diet for the growing children and expectant mothers. Milk, is a major constituent of the diet, its quality assurance is considered essential to the welfare of a community. Milk is nutritious food for human beings, also serves as a good medium for the growth of many microorganisms, especially *Lactobacillus, Streptococcus, Staphylococcus and Micrococcus sp.* Bacterial contamination of raw milk can originate from different sources from animals such as air, milking equipment, feed, soil, feces and grass (Torkar & Teger, 2008). Milk micorflora includes spoilage and pathogenic microorganisms. Many milk borne diseases such as tuberculosis, brucellosis and typhoid fever are known (*Goff and Horst*, 1995). Milk is spoiled by a wide range of microorganisms some of which are pathogenic and are responsible for milk borne diseases. The milk is very easily contaminated if collected unhygeinically and handled carelessly leading to quick spoilage (prajapati, 1995, chatterjee *et.al.,* 2006) and is often contaminated by *Escherichia coli* bacteria under poor sanitary conditions which can affect public health. The coliform group of bacteria is defined as the indicator (faecal coliform) of suitability of milk for consumption (Standard method committee, 1981).

MATERIALS AND METHODS

240 raw milk and 72 pasteurized milk samples were collected form diverse locations of Warangal district and surrounding villages in sterile screw cap tubes. After collection, the samples were transported to the laboratory on ice in sterile condition and processed for MBRT and coli form test with in three hours.

In the methylence blue reduction (MBRT) test 1 ml of methylene blue (1:25,000) is added to 10ml of milk. The tube is sealed with rubber stopper and slowly inverted three times to mix. It is placed in a water bath at 35^{o}C and examined at intervals up to 6hrs. The time taken for the methylene blue to become colorless is the methylene blue reduction time (MBRT). The grading of milk sample's on the basis of methylene blue reduction test in different milk samples are presented Table. (Benson, 2002). The methylene blue reduction test depends upon the ability of bacteria in milk to grow and to consume the dissolved oxygen, which reduces the oxidation reduction potentials in the medium.

Isolation of Microorganisms from milk samples: Serial dilutions of samples were made up to 10^{-6} in nutrient broth and Mac Conkey broth. Samples were plated in duplicate using pour plate technique. 0.5ml of the diluted sample was delivered by pipette in to 19.5 ml of enriched agar. Plates were inverted in an incubator at 37^{o}C for 24-48hrs. Total viable counts were carried out on nutrient agar. Quantitative analysis for the presence or absence of specific microorganisms was done by plating on selective media.

Characterization of isolates from milk samples: At intervals, colonies on the incubated plates were picked and purified by repeated sub-culturing by streaking on the desired media with a sterile wire loop. The strategy consisted of picking one colony to represent every visibly different morphology on each plate. A maximum of 5 colonies were obtained per sample, which were examined microscopically for Gram's reaction and colony morphology (shape, size, colour, texture) using 24h old cultures. Motility and biochemical tests were performed .Appropriate positive and negative controls

were used to make distinction positive and false-positive reactions

***Identification of isolates from milk samples*:** Identification was based on growth on selective agar and broth, colony morphology, Gram's reaction, biochemical test results and criteria for disregarding negative cultures. Results were analyzed using Bergy's manual, and other methods for the identification (Barrow &Feltham, 1993; De Silva *et al.,* 2001; Ellis&Goodacre, 2006).

RESULTS AND DISCUSSION

The results are presented in tables 1 and 2. A critical perusal of the table 1 reveals that out of 240 raw milk samples tested, 46 (19.1%) samples were found to be good, 61 (25.4%) samples were fair. The highest numbers of samples were found to be poor 65 (27.0.%) and very poor 68 (28.3%). During this study it was found that in May and June most samples were very poor or poor and this may be due to high temperature prevailing in summer reason. Out of 72 pasteurized milk samples, highest number of samples were found to be good 59 (81.9%) only 3 (4.1%) samples were found to be very poor, 4 (5.5%) samples were poor. Chatterjee *et.al.,* (2006) reported that the raw milk contained higher number of micro flora probably due to contamination from the animal. Bacteria found in manure, soil and water may enter milk due to dairy utensils and milk contact surfaces. Present study showed that 53% and 49% of raw milk samples were of very poor & poor category but in case of pasteurized milk samples, 86% of the samples were of good quality due to pasteurization. The study indicated that the dominant microbial flora associated with poor milk samples in and around Warangal District, (A.P.) were in the order of *Lactobacilli, Staphylococcus aures, Escherichia coli, Bacillus subtilis, Salmonella typhi, Faecal coliforms.* In pasteurized milk samples also very poor quality were recorded only in May and June months.

TABLE 1
MICROBIOLOGICAL QUALITY OF CAN/POT MILK SUPPLIED TO WARANGAL CITY (A.P)

Month	Total No. of samples	Quality of milk			
		Very poor	Poor	Fair	Good
January	40	5(12.5%)	7(17.5%)	14(35.0%)	14(35.0%)
February	40	7(17.5%)	9(22.5%)	13(32.5%)	11(27.5%)
March	40	9 (22.5%)	10(25.0%)	12(30.0%)	9(22.5%)
April	40	13(32.5%)	13(32.5%)	10(25.0%)	4(10.0%)
May	40	15 (37.5%)	15(37.5%)	5(12.5%)	5(12.5%)
June	40	19 (47.5%)	11(27.5%)	7(17.5%)	3(7.5%)
Total	**240**	**68 (28.3%)**	**65 (27.0%)**	**61 (25.4%)**	**46 (19.1)**

TABLE 2
MICROBIOLOGICAL QUALITY OF PASTEURIZED MILK SUPPLIED TO WARANGAL CITY (A.P)

Month	Total No. of samples	Quality of milk			
		Very poor	Poor	Fair	Good
January	12	0(0%)	0(0%)	2(16.6%)	10(83.3%)
February	12	0(0%)	0(0%)	1(8.3%)	11(91.6%)
March	12	0(0%)	0(0%)	2(16.6%)	10(83.3%)
April	12	0(0%)	0(0%)	0(0%)	12(100%)
May	12	1(8.3%)	2(16.6%)	0(0%)	9(22.5%)
June	12	2(16.6%)	2(16.6%)	1(8.3%)	7 (58.3%)
Total	**72**	**3 (4.1%)**	**4 (5.5%)**	**6 (8.3%)**	**59 (81.9%)**

Table 3
BACTERIA PRESENT IN RAW MILK SAMPLES SUPPLIED TO WARANGAL CITY (A.P)

Month	Total no. of samples	Number of colonies appeared					
		Lactobacilli	*Staphyloco-ccus aureus*	*Escheriti a coli*	*Bacillus subtilis*	*Salmone-lla typhi*	*Fecal coliforms*
January	40	62	42	70	12	-	2
February	40	56	40	20	11	2	-
March	40	48	36	21	14	3	2
April	40	50	48	26	13	-	-
May	40	72	58	35	20	5	4
June	40	86	62	39	25	6	4
Total	**240**	**374**	**286**	**158**	**95**	**16**	**8**

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