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# An Awareness Study on Human Milk Banks and Microbial Analysis of Human Milk

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**Abstract:** Human breast milk is considered to be the most important nutrient for new born babies, especially preterm ones. If the mother does not produce sufficient milk, fresh frozen un pasteurized milk from a milk bank should be available to all ill neonates until the mother's own production is established. Bank milk should therefore be free of pathogenic bacteria and generally have a bacterial content of less than 104 colony forming units/ml. The donors should test negative to cytomegalovirus (CMV), HIV, and hepatitis B and C, in order to prevent a serious viral infection. Early use of breast milk for preterm infants has resulted in a reduced incidence of necrotizing enterocolitis, faster tolerance of enteral feeding, and thus a reduced need for parenteral nutrition. The present study was aimed to analyze the microbial analysis of Human milk collected from Human milk bank of Niloufer Hospital, Red hills. The milk sample was collected in glass sterile bottle, which had been pasteurized. The sample was analyzed using Most Probable Number(MPN) method to detect the presence of coli forms in human milk. The result of the study revealed was 0.03/10ml, which indicated that the milk was less contaminated and safe for infant consumption. A survey on 100 lactating women was also done to make them aware about the Human milk banks. A well-structured questionnaire was employed to interview the subjects about donating breast milk, importance of colostrum and to assess their opinion on utilization of milk from such banks for their neonates. Thus, it was concluded from our study that the majority of women didn't want to donate breast milk to human milk banks. It was also concluded the storage, handling procedure and proper pasteurization temperature of milk in human milk bank minimizes the milk contamination. proper pasteurization temperature, storage and handling process of milk in human milk banks may minimize the microbial contamination.

## I. INTRODUCTION

Human breast milk is considered to be the most important nutrient for new born babies, especially preterm ones [1]. Breastfeeding is the best method of infant feeding because human milk continues to be the only milk which is tailor-made and uniquely suited to the human infant. All mothers should be encouraged to breast-feed their infants [2]. If the mother does not produce sufficient milk, fresh frozen pasteurized milk from a milk bank should be available to all ill neonates until the mother's own production is established [1,2,9]. Human milk provides health benefits for all new born infants but is of particular importance for high-risk infants, especially those born with very low birth weight. Donor human milk also can be beneficial to supplement the mother's own milk when necessary [3]. Bank milk should therefore be free of pathogenic bacteria and generally have a bacterial content of less than 104 colony forming units/ml. The donors should test negative to cytomegalovirus (CMV), HIV, and hepatitis B and C, in order to prevent a serious viral infection. Early use of breast milk for preterm infants has resulted in a reduced incidence of necrotizing enterocolitis, faster tolerance of enteral feeding, and thus a reduced need for parenteral nutrition [1,4]. It is universally accepted that breast milk is the optimum exclusive source of nutrition for the first six months of life and may remain part of the healthy infant diet for the first two years of life and beyond. Human milk is species specific and is thus, markedly superior to all alternatives for new born feeding. Although bovine- and plant-based formulas approach the fat, protein and carbohydrate composition of human milk, they are not able to replicate the complexity or functionality of other bioactive factors found in human breast milk. The benefits of human breast milk include optimum growth, immune function and development at minimal cost to the family [3]. Breast milk is the normal way to feed infants; it is accepted worldwide as the optimal exclusive first source of nutrition. Human milk is recognized for its numerous benefits including inducing tolerance to allergens, providing passive immunization, improving lipid profiles, and controlling blood pressure [5]. Human colostrum (HC) is the first milk produced after birth and is particularly rich in immunoglobulin (Ig), antimicrobial peptides (lactoferrin and lacto peroxidase) and other bioactive molecule, including growth factors which are important for nutrition, growth and development of new born infants and also for passive immunity [6]. Premature infants constitute the largest and most important group of infants where milk from other women is needed because their own mothers' milk is not available or is not available in sufficient quantity. Milk from mothers who deliver prematurely (preterm milk) has been shown to be different from milk of mothers who deliver at term (term milk). Preterm milk has been noted to have increased

amount of nitrogen, total protein, immune proteins, total lipids, medium chain fatty acids, total energy, some vitamins and minerals as well as trace elements. The long chain polyunsaturated fatty acids (especially docosahexaenoic acid and arachidonic acid) found in both term and preterm milk have been implicated in optimal brain development and retinal maturation [7].

Human milk banks collect, screen, pasteurize, and distribute donated breast milk to hospitals or outpatient recipients [8]. A study was done to determine the variability in fatty acid and amino acid composition in individual and pooled PDM using current pooling practices in place at milk banks compliant with Human Milk Banking Association of North America (HMBANA) guidelines, specifically for protein (0.7–1.0 g/100 mL) and caloric contents (67–81 kcal/100 mL) [9].

Human milk banks collect, screen, pasteurize, and distribute donated breast milk to hospitals or outpatient recipients [8]. Storage should be done in the same container that is used for pasteurization.

It is advisable not to transfer processed milk in other containers as it has risk of contamination. Culture negative processed milk should be kept at -20°C in tightly sealed container with clear mention of expiry date and other relevant data on the label.

It can be preserved for 3 to 6 months. Random cultures of preserved milk before disbursement can aid quality assurance [2].

## II. MATERIALS AND METHOD

An analytical, descriptive and stratified study was performed with a sample of 100 subjects, which included lactating mothers. Data was collected from lactating women by visiting maternity hospitals in Hyderabad like, Modern Maternity Government Hospital-Petla burj, Nisa Maternity Hospital-Red Hills, Amba Hospital- Bazar Ghat.

An interview cum questionnaire method was used to collect data from the respondents, who were questioned about their lifestyle, general information and information about Human Milk Banks. Microbial analysis of human milk is also done by collecting milk from Human Milk Bank situated at Niloufer Hospital-Red Hills, Hyderabad.

The microbial analysis of milk was done by using biochemical tests such as MPN (Most probable Number or Multiple tube Fermentation test) presumptive test, confirmatory test and complete test, triple sugar iron test (TSIA), gram staining was adapted to detect the Coliform Bacteria in the sample.

## III. RESULTS AND DISCUSSION

### A. Colony Morphology

It is done for microbial growth on the media. The process was carried out using MPN method. The human milk collected from human milk bank shows 0.03/10ml coli forms, which indicated the milk is less contaminated and safe for infant consumption.

The sample was taken for further test which was confirmatory and complete in which formation of metallic green sheen had shown which detected the presence of E-coli. The presence of bacteria in human milk may be due to storage and handling procedure or during the process of transportation.

### B. Presumptive Test

This test is specific for the detection of coliform bacteria. In this test MacConkey broth is used as media. Coliform bacteria capable of this broth as carbon source doesn't produce gas or acid. Which indicated it is safe for infant consumption.

### C. Confirmatory Test

In this test positive MacConkey tubes were gently agitated and loopful of suspension transfer to tube of brilliant green bile broth. Gas formation in Durham's tubes regarded as positive confirmatory test.

### D. Complete Test

In this test Levin Eosin methylene blue (L-EMB) agar was used. The plates were streaked with a loopful of suspension from confirmed positive Brilliant Green Bile Broth Culture Coliform colonies were form on EMB Agar. Then the reconfirmation of coli form is done by inoculating the colonies in MacConkey broth and brilliant bile broth at 24 hours and by streak it on triple sugar iron agar tube after streaking of plates the colony culture was incubated at 35C for 24 hours and prepared gram stained slide from the plate and the gram negative non-spore rods have seen and the complete test is resulted as confirmed positive test.

percentage of women aware of milk donating banks

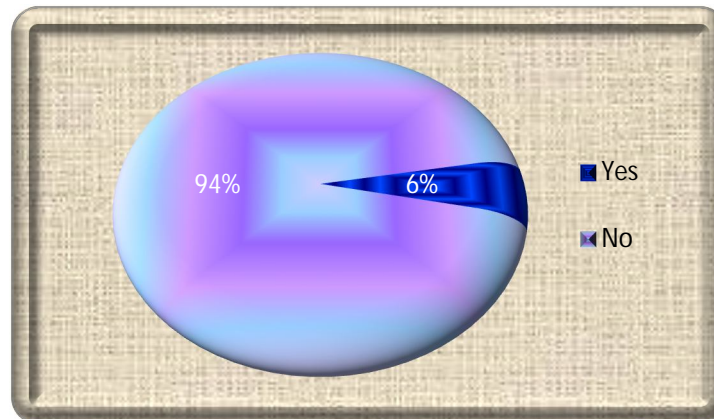


Fig. 1 shows that out of 100 subjects majority of the subjects are not heard of milk donating banks 94% and only 6% of the subjects heard of milk donating banks.

percentage of women interested in donating milk to human milk banks

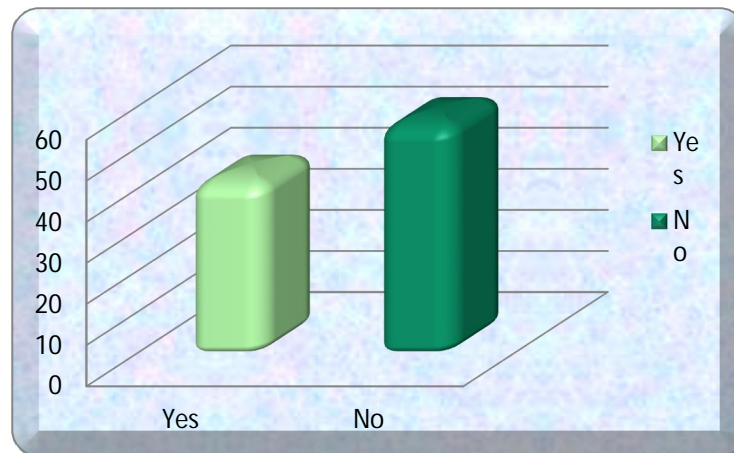


Fig. 2 shows that out of 100 subjects, 43% of the subjects want to donate breast milk to banks and 57% of the subjects do not want to donate breast milk to banks.

percentage of women interested in utilizing milk from human milk banks

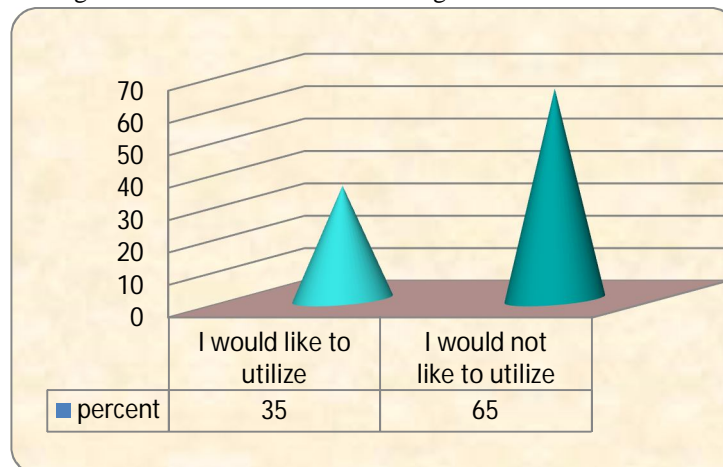


Fig. 3 shows that out of 100 subjects, 35% of the subjects like to utilize milk banking and 65% of the subjects do not like to utilize milk banking at the time of hindering.

Percentage of women aware about the importance of colostrum

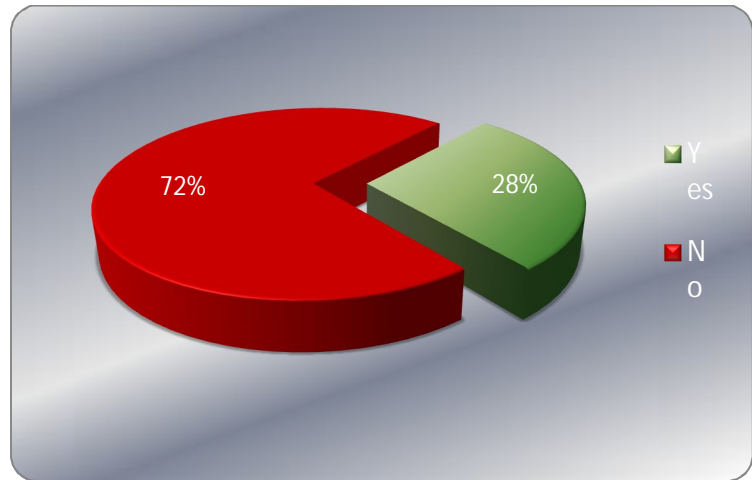
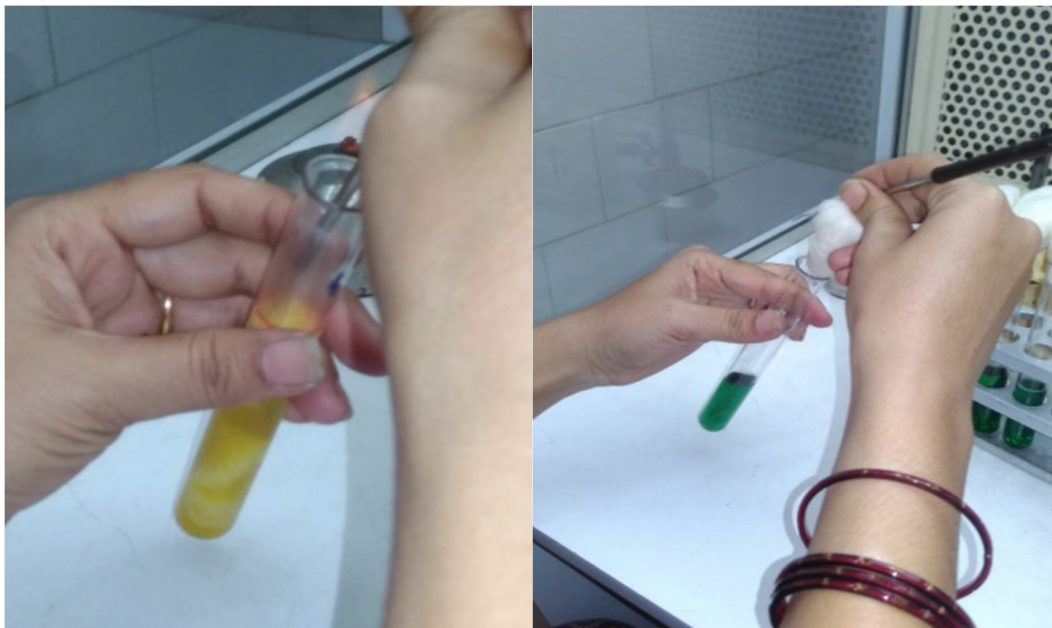


Fig. 4 shows that only 28% of the subjects know the importance colostrum/first milk and the remaining of the subjects do not know.



Pipetting the Milk sample in MacConkey broth Gas bubble observed



Loopful of suspension transferred



EMB Agar plate



Streaked plate



EMB Agar Plate with Coli Form Colonies

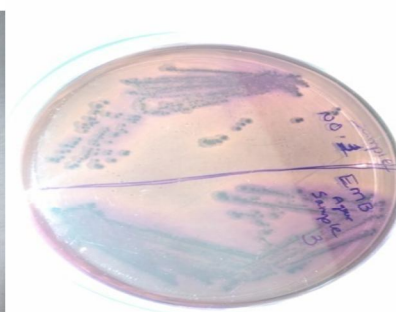
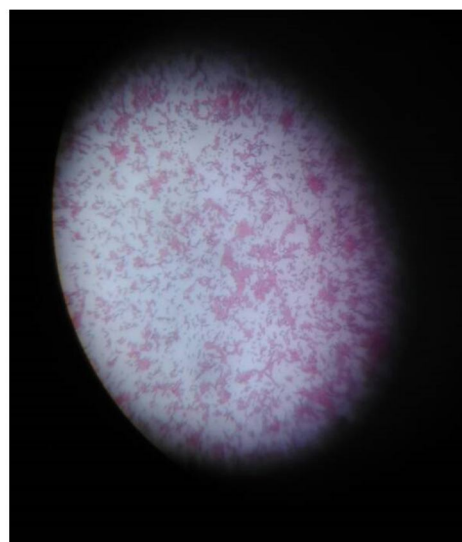


Plate with metallic green sheen colonies



Coli form colonies seen under microscope



Picture showing non-spore rod shaped gram-negative bacteria.

#### IV. CONCLUSION

From the above graphical and microbial analysis, our study concluded that the storage, handling procedure, proper pasteurization temperature of milk in human milk bank minimizes the milk contamination. It also concluded that majority of women didn't want to donate breast milk to human milk banks and majority of women are not aware about the importance of colostrum.



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