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Bacteriological Profile and Its Antimicrobial Susceptibility Pattern of Top Feeding Bottles from a Tertiary Care Centre

Dr.P.Tejaswi Sai[1], Dr.V.Sudha Rani[2], Dr.G.Jyothi Lakshmi[3]Dr.P.ShashikalaReddy[4]

[1](Post Graduate, Department of Microbiology, Osmania Medical College, Koti, Hyderabad.)
[2](Professor, Department of Microbiology, Osmania Medical College, Koti, Hyderabad.)
[3](Professor & HOD, Department of Microbiology, Osmania Medical College, Koti, Hyderabad.)
[4](Principal & Professor, Department of Microbiology, Osmania Medical College, Koti, Hyderabad.)

Abstract:
Background:*In India each year two- thirds of infant deaths occur due to inappropriate feeding practices. Feeding bottles with improper decontamination can lead to gross contaminationwith enteric pathogens, resultant gastroenteritis from hygiene malpractices can be extremely dangerous. Therefore, implementation of effective cleaning and disinfection procedures for feeding bottles is extremely important.*
Materials andMethods:*50 Feeding bottles were collected from babies within 4 hours of use, sterile swabs were used to obtain swipe from 4 sites of feeding bottles and inoculated onto Blood agar and Mac Conkey agar plates and incubated at 37^{0}c for 24 hours. Isolated organisms identified by standard bacteriological procedures and antimicrobial susceptibility testing of the isolates done.*
Results: *Out of 50 bottles processed, isolates were obtained from all the bottles. Among these isolates, Klebsiella species were46% (23), Coagulase negative Staphylococciwere28% (14), Enterococcus specieswere 12% (6), Pseudomonas species were 10% (5), Staphylococcus aureus were 4% (2), and all isolates showed resistance pattern of varying degree to commonly used antibiotics.*
Conclusion:*To conclude this study shows that isolates were obtained from all the feeding bottles. Therefore, either effective decontamination of feeding bottles or use of alternatives like spoon, katori is essential to decrease infant mortality related to inappropriate feeding practices.*
Key words:*Feeding bottles, decontamination, alternative feeding practices.*

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I. Introduction

Breastfeeding has been declined in the recent years due to empowerment of women. It provides complete nutrition for the baby and is influenced by many factors such as demographic, biophysical, social, cultural and psychological factors. Eachyear more than 2.4 million infant deaths occur in India and two thirds of these are because of inappropriate feeding practices.[1] WHO recommends early initiation of breast feeding within an hour of birth, exclusive breast feeding till 6 months of age, complementary feeds should be introduced at 6 months with continuing breast feeding up to 2 years or beyond.[2] Cup or palladi which is a cup with beak or spoon should be used to feed low birth weight infants who require an alternative oral feeding methods.[3]

Breast feeding is beneficial to both infant and mother. It reduces infections and mortality among infants, improves mental and motor development and protects against obesity, metabolic diseases, cardiovascular risk factors and allergy.[4,5]

Contaminated ingredients and water, as well as inadequate practices involved in preparation, washing and disinfection of utensils used to prepare feeding bottles are traditionally considered as risk factors for contamination of even domestically prepared feeding bottles.[6,7]Infant formula powder may also be unsterile and may contain coliforms and other bacteria in low number, this milk may constitute an excellent medium for bacterial growth, hence the preparation, handling and duration of storage is also important.[8]Contamination may also be due to transfer of bacteria from personnel which depends on load of bacteria and its survival on the hands.[9]

Bacterial infection in infants when exposed to contaminated foods may be because they have developmentally premature immune system and gastrointestinal tract.[9]Bottle fed baby has 2 to 3 times higher death rate and frequent episodes of diarrhea and respiratory illness than exclusively breast-fed counter parts.[10]

High levels of coliforms detected in bottles can cause biofilm formation on interior walls of bottles and cannot be removed by gentle rinsing, therefore more intensive disinfection methods are needed to maintain

clean bottles.[11] The present study is carried out to know the bacteriological contamination among top feeding bottles and to create awareness of breast feeding among care givers.

II. Materials & Methods

The prospective, cross sectional study was carried out on babies with top feeding bottles inNilouferHospital, Hyderabad, Telangana from October 2019 – December 2019. Institutional Ethics Committee approval was obtained prior to the study.

Study Design – Prospective cross-sectional study.

S**tudy Location**: Department of Microbiology, Osmania Medical College and Department of Paediatrics, Niloufer hospital Hyderabad.

Study Duration- October 2019 to December 2019 (3 months duration)

Sample size – 50 top feeding bottles.

Inclusion criteria:

1. Both male and female babies
2. Age group from day zero to 2 years
3. Babies with top feeding bottles admitted in Niloufer hospital
4. Parents willing to give consent.

Exclusion criteria-

1. Age more than 2 years
2. Unused and washed bottles
3. Parents not willing to give consent.

Procedure methodology –

Consent was obtained from those who were willing to participate in the study. Feeding bottles were collected from bottle fed babies admitted in the wards within four hours of use after washing hands with soap and water and transported to Microbiology laboratory at ambient temperature and processed within 2 hours for microbiology assay.

Sterile swabs were used to obtain swipe from feeding bottles and each feeding bottle was sampled in 4 locations – 1. Inner side of nipple, 2. Inner rim of cap, 3. Interior of bottle from middle, 4. Interior of bottle from bottom.

Swabs were inoculated onto Blood agar and MacConkey agar plates and incubated aerobically at 37^0c for 16 to 18 hours and plates were observed for growth.

Identification of bacterial pathogens were done based on colony morphology, staining characteristics, cultural and biochemical properties using standard laboratory techniques.

Antibiotic susceptibility testing was done for all the isolates on Mueller Hinton agar by Kirby Bauer disc diffusion method by using commercially available Hi media antibiotic discs as per CLSI guidelines.

Table no 1 shows list of antibiotics used for antibiotic susceptibility testing on Mueller Hinton agar.

Table no 1: List of antibiotics

Gram Negative organisms		**Enterococcus**		**Staphylococcus aureus and CoNS**	
Antibiotics	**Concentration (mcg)**	**Antibiotics**	**Concentration (mcg)**	**Antibiotics**	**Concentration (mcg)**
Ampicillin	10	Ampicillin	10	Gentamicin	10
Ciprofloxacin	5	High level Gentamicin	120	Ciprofloxacin	5
Ceftriaxone	30	Vancomycin	30	Cefoxitin	30
Cefepime	30	Linezolid	30	Clindamycin	2
Piperacillin/Tazobactam	30/6	Azithromycin	15	Cotrimoxazole	25
Imipenem	10	Doxycycline	30	Vancomycin	30
				Linezolid	30

III. Results

Total number of culture positives - 50

Out of 50 culture positives

Gram negative bacteria - 28 (56%)

Gram positive bacteria - 22(44%)

Figure no 1 shows percentage of bacterial isolates of Feeding bottles.

Figure no 1: Bacterial Isolates of Feeding Bottles

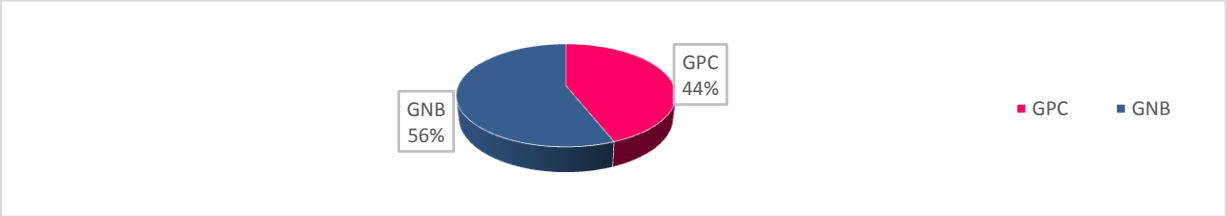

Figure no 2 shows age wise distribution of samples, 18% babies were initiated on bottle feeding within 10 days of birth, 14% during 11 days to 1 month, 8% during 2 to 4 months, 32% during 5 to 8 months of age,18% during 9 to 15 months, 10% during 16 to 24 months.

Figure no 2: Age wise distribution of samples

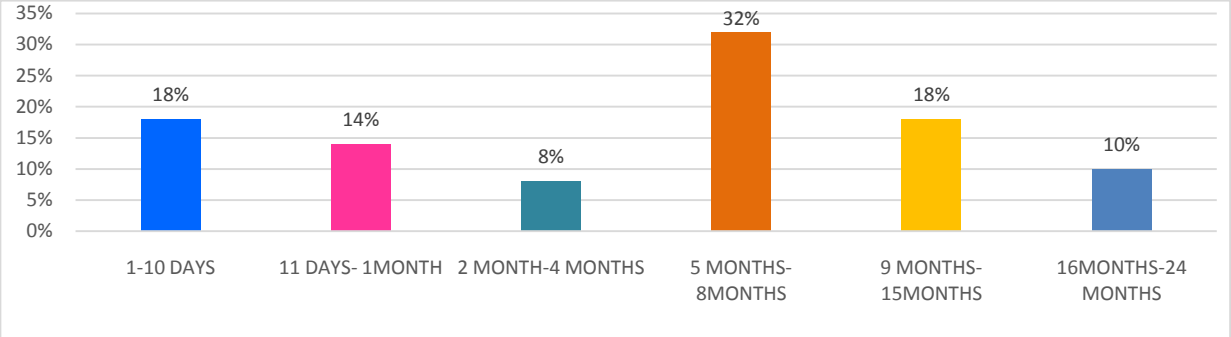

Table no 2 shows Bacterial isolates, most common isolate found was Klebsiella species 46% (23), followed by CoNS 28% (14), Enterococcus species 12% (6), Pseudomonas aeruginosa 10% (5) and Staphylococcus aureus 4% (2).

Table no 2: Bacterial Isolates

S.No	CULTURE SMEAR RESULT	NUMBER	PERCENTAGE(%)
1	Klebsiella species	23	46
2	CoNS	14	28
3	Enterococcus species	6	12
4	Pseudomonas aeruginosa	5	10
5	Staphylococcus aureus	2	4

Table no 3 shows sensitivity pattern of Gram-negativebacilli. They were 100% sensitive to Imipenem and Ciprofloxacin, followed by 96.4% sensitive to Piperacillin-Tazobactam followed by Cefepime, Ceftriaxone and Ampicillin.

Table no 3: Antimicrobial susceptibility pattern of Gram-Negative Bacilli

ANTIBIOTIC	SENSITIVITY(%)(No of isolates)	RESISTANCE(%)(No of isolates)
Imipenem (10 mcg)	100 (28)	NIL (0)
Ciprofloxacin (5 mcg)	100 (28)	NIL (0)
Piperacillin-Tazobactam (30/6 mcg)	96.4 (27)	3.6 (1)
Cefepime (30 mcg)	71.4 (20)	28.6 (8)
Ceftriaxone (30 mcg)	10.7 (3)	89.3 (25)
Ampicillin (10 mcg)	10.7 (3)	89.3 (25)

Table no 4 shows sensitivity pattern of Staphylococcus aureus and CoNS. They were 100% sensitive to Ciprofloxacin, Gentamicin, Vancomycin, Linezolid followed by 62.5% sensitive to Cotrimoxazole followed by Cefoxitin and Clindamycin.

Table no 4: Antimicrobial Susceptibility pattern ofCoNS and Staphylococcus aureus

ANTIBIOTIC	SENSITIVITY(%)(NO of isolates)	RESISTANCE (%)(No of isolates)
Ciprofloxacin (5 mcg)	100 (16)	NIL (0)
Gentamicin (10 mcg)	100 (16)	NIL (0)
Vancomycin (30 mcg)	100 (16)	NIL (0)
Linezolid (30 mcg)	100 (16)	NIL (0)
Cotrimoxazole (23.75/1.25 mcg)	62.5 (10)	37.5 (6)
Cefoxitin (30 mcg)	43.7 (7)	56.3 (9)
Clindamycin (2 mcg)	6.2 (1)	93.8 (15)

Table no 5 shows sensitivity pattern of Enterococcus. It was 100% sensitive to Vancomycin, High level Gentamicin, Azithromycin, Linezolid followed by 66.7% sensitive to Doxycycline and Ampicillin.

Table no 5: Antimicrobial Susceptibility pattern of Enterococcus species

ANTIBIOTIC	SENSITIVITY(%) (No of isolates)	RESISTANCE(%) (No of isolates)
Vancomycin (30 mcg)	100 (6)	NIL (0)
High level Gentamicin (120 mcg)	100 (6)	NIL (0)
Azithromycin (15 mcg)	100 (6)	NIL (0)
Linezolid (30 mcg)	100 (6)	NIL (0)
Doxycycline (30 mcg)	66.7 (4)	33.3 (2)
Ampicillin (10 mcg)	66.7 (4)	33.3 (2)

IV. Discussion

Bacterial isolates were obtained from all feeding bottles. The organisms isolated were mainly gram negative, among these Klebsiella was most common 46% (23), followed by Pseudomonas 10% (5). Among, Gram positive organisms most common was Coagulase negative Staphylococcus 28% (14), followed by Enterococcus 12% (6), Staphylococcus aureus 4% (2).

In the present study Enterobacteriaceae accounted for 46% whereas in a study done by Chandra Sekharkondapalli et al,[1] it accounted for 69% and 15% in Elizabeth C Redmond et al[12]. In the present studyStaphylococcus aureus accounted for 4% whereas in study done by ChandraSekharkondapalli et al[1]it accounted for 16% and 12% in Elizabeth C Redmond et al[12].

Gram negative organisms showed sensitivity to Imipenem, Piperacillin- Tazobactam, Ciprofloxacin. Coagulase negative Staphylococcus and Staphylococcal aureus showed sensitivity to Vancomycin, Linezolid, Ciprofloxacin, Gentamicin in the present study.

Staphylococcus was 100% sensitive to Vancomycin and Linezolid in our study which was correlated to Chandra SekharKondapalli et al[1].Pseudomonas was resistant to Ampicillin in our study which was correlated to Chandra SekharKondapalli et al[1].CoNS was 100% sensitive to Vancomycin and Linezolid in our study which was correlated to Chandra SekharKondapalli et al[1].

In the present study, babies were admitted mostly with respiratory symptoms which accounted for 42%, followed by CNS symptoms 23%, gastrointestinal symptoms 19% and sepsis 16%, where as in a study done by Chandra Sekharkondapalli et al[1] gastrointestinal symptoms accounted for 43%, followed by respiratory symptoms 39% and other illness 18%.

In the study by Sarah Gibson et al[13] the predominant practice of cleaning the bottles was by rinsing with tap water or warm water which was inadequate and the screw tops can be particularly difficult to clean, same practices were followed in our study.

Contamination is probably a multifactorial in origin. As human skin acts as reservoir of pathogens and cross contamination may occur, use of non-sterile disposable gloves and use of effective decontamination procedures for feeding bottles which include use of clean, hot water and detergent followed by rinsing will be beneficial.[12,14]

V. Conclusion

The main cause of weaning of breast milk feeding and shift over to bottle feeding is due to lack of awareness. Those babies who are initiated with bottle feeding before the age of 6 months had frequent illness. So, parents should be educated about the risk of bacterial contamination of feeding bottles and should be encouraged for exclusive breastfeeding or use of better alternative feeding practices like feeding with spoon or katori, and if necessary, use of proper precautions like hand hygiene, environmental measures and use of effective decontamination procedures for feeding bottles.

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