

Original Research Article

Bacterial contamination of nasogastric feeding tube and development of neonatal sepsis in premature newborns: a prospective observational research at a tertiary care center in Gujarat, India

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ABSTRACT

Background: The objective of the present study is to observe the effect of frequency of changing nasogastric feeding tube (NG-FT) on microbial growth in relation to development of neonatal sepsis in premature newborns. The study is prospective observational study in its nature. Neonatal intensive care unit (NICU) of a tertiary care teaching hospital of western Gujarat, India, from December 2016 to November 2017.

Methods: Eighty-five preterm newborns admitted to NICU for feeding support were randomly divided into three groups depending on frequency of changing nasogastric feeding tubes (Group I, II and III with NG-FT changed every 12, 24 and 48 hourly respectively). In Groups I, II and III, the first NG-FT cultures were sent at the end of 12, 24, 48 hours of NG-FT insertion respectively. The second and third NG-FT cultures were sent after 7 and 14 days respectively. Microbial growth pattern was observed and correlated with development of necrotizing enterocolitis (NEC), neonatal sepsis and mortality.

Results: Microbial growth on first NG-FT culture significantly increased when frequency of changing nasogastric feeding tube was reduced from every 12 to 24 or 48 hours { $p = 0.0432$ (Group I vs II) and 0.0001 (Group I vs III)}. Microbial growth increased on second (24.1%, 87% and 85% in groups I, II and III respectively) and third NG-FT culture (44% in group I and 100% in groups II and III); this was significantly high in group II and III as compared to group I ($p = 0.0001$). Common organisms isolated were *Klebsiella pneumonia* (63.28%), *Pseudomonas aeruginosa* (32%) and *Escherichia coli* (21%). No significant difference was noted in incidences of feeding intolerance, necrotizing enterocolitis (NEC), neonatal sepsis and mortality among the three groups.

Conclusions: Organism growth in nasogastric feeding tube culture increases significantly when frequency of changing NG-FT is beyond 12 hours. However, there is no increase in episodes of feeding intolerance, necrotizing enterocolitis (NEC), neonatal sepsis and mortality.

Keywords: Antibiotic sensitivity, Nasogastric feeding tube culture, Neonatal sepsis, Organisms, Preterm new borns

INTRODUCTION

Preterm newborns admitted to neonatal intensive care unit (NICU) for gavage feeding assistance are vulnerable to acquire infection. It has been documented that enteral delivery systems such as nasogastric feeding tubes are susceptible to bacterial growth.¹ Since the tubes are at

body temperature and contain nutrients (breast milk or infant formula), it is reasonable to anticipate that bacteria will multiply in the tubes and grow as bacterial biofilm.

The biofilm will inoculate subsequent routine feeds and may survive passage through the neonate's stomach.^{1,2} Premature neonates have a low immune status and lack a

competing intestinal bacterial flora. Hence these biofilms associated organisms could result in neonatal infections.³ Neonatal sepsis is one of the commonest causes of neonatal mortality contributing to significant (19%) of neonatal deaths.⁴ External factors such as environment and prolonged use of a feeding tube affect the colonization of gut. There is association between gut colonization and neonatal sepsis.⁵ This prospective interventional study was done to know the effect of frequency of changing feeding tube on microbial growth, its antibiotic sensitivity and its correlation with development of feeding intolerance, necrotizing enterocolitis (NEC), neonatal sepsis and mortality.

METHODS

This prospective observational study was conducted in a tertiary care NICU from December 2016 to November 2017. The study was approved by institutional ethics committee. The written and informed consent of the parents was taken.

All preterm newborns admitted in NICU of our institute for gavage feeding support only were included in the present work. Preterm newborns with antenatal risk factors for sepsis, evidence of sepsis clinically or by sepsis screening test and those with history of resuscitation at birth were excluded. Antenatal risk factors considered for sepsis were prolonged rupture of membrane > 24 hours prior to delivery (PROM), foul smelling liquor, maternal fever within 2 weeks of delivery or during labor, prolonged labor and >3 sterile or single unclean per vaginal examinations.⁶

Sepsis screening tests included estimation of C-reactive protein (> 1 mg/dl), total leukocyte count (< 5000/mm³ or > 25000/mm³), micro-ESR (> 15 mm in 1st hour), absolute neutrophil count: low counts (as per Manroe chart for term and Mouzinho's chart for VLBW infants) and immature/total neutrophil ratio (> 0.2).⁷⁻¹⁰ Those patients who met inclusion criterion were divided into three groups (I, II and III) by computer generated random numbers. Nasogastric feeding tube (NG-FT) was changed every 12, 24 and 48 hours in Groups I, II and III respectively. In Groups I, II and III, the first NG-FT cultures were sent after 12, 24, 48 hours of NG-FT insertion respectively.

The second and third NG-FT cultures were sent after 7th and 14th day of NG feeding respectively. To send the cultures, NG-FT was cut and its distal end which remained inside the body was sent in sterile container to microbiology laboratory. It was immediately put into liquid brain heart infusion broth and incubated overnight at 37° C. Next day subculture was done from inoculated broth onto blood/MacConkey agar and incubated at 37° C in aerobic conditions. Isolates from inoculated plates were identified by standard microbiology techniques. NG-FT was defined as contaminated if organism growth was > 10³ Colony Forming Units (CFU). Antibiotic

sensitivity was performed by disc diffusion method as per CLSI guidelines.¹¹ We correlated the results of NG-FT culture with development of sepsis clinically or by sepsis screening if needed. Confirmation of sepsis was done by standard blood culture. Lumbar puncture for cerebrospinal fluid examination was done in all cases prior to starting antibiotics.¹² Primary objective of this study was to determine the effect of frequency of changing NG-FT on microbial growth. Secondary objective was to know the correlation between frequency of changing NG-FT and development of NEC as per modified Bell's staging, sepsis and mortality in all three groups.¹³

Feeding protocol: Under aseptic precautions, nursing staff or residents of our NICU were trained to insert NG-FT and changed it according to random allocation of groups. Initially, feeding was started at 20-30 ml/kg/day at 2 hourly intervals and then increased up to 40-50 and 60-80 ml/kg/day after 12 and 24 hours of life respectively.

On third day, human milk fortifiers (HMF) was added to expressed breast milk (EBM) when 100 ml/kg/day feeds were reached. Full feeds were defined at 180 ml/kg/day. Infant powder formula (IPF) was added if EBM was not sufficient or available. When the signs of feed intolerance, NEC or sepsis were noticed, the feed was withheld and again restarted when these signs resolved. Patients were discharged after being shifted to katorispoon feed/ breast feeding, gaining weight at 10-20 gm/kg/day for at least three consecutive days and when weight was > 1.5 kg. Follow up was done at 1st week, 2nd weeks and 1st month after discharge. All the data was tabulated in Microsoft excel sheets and statistical analysis was done using ANOVA test for continuous variable and Chi-Square test for categorical variable. Significance was defined as p value was < 0.05.

RESULTS

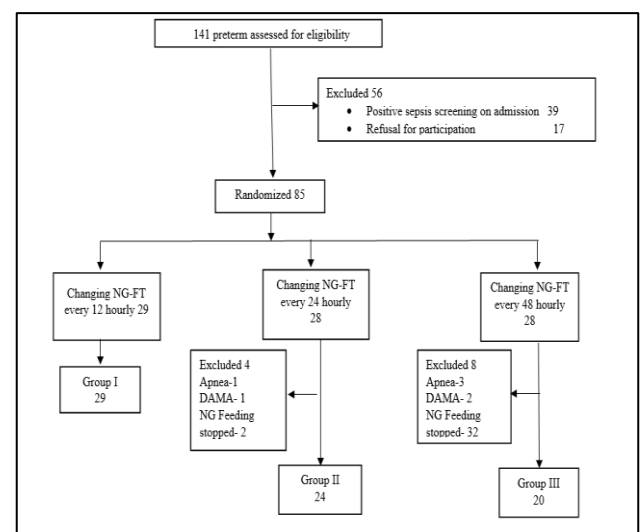


Figure 1: Flowchart of study participants.

141 preterm neonates were admitted in NICU for nasogastric feeding during the study period, of which 85 fulfilled inclusion criteria (Figure 1). The baseline

characteristics were comparable in all three groups. Time to reach full feeds (p = 0.659) and duration of hospital stay (p = 0.574) were also similar in all groups (Table 1).

Table 1: Demographic profile of patients.

Characteristics	Group I (n=29) ^{\$}	Group II (n=24) ^{##}	Group III (n=20) ^{\$\$}	p value
Gestational age (weeks)*	33.17 (1.4)	32.79 (1.8)	33.25 (1.8)	0.619
Male:Female [#]	18:11	10:14	8:12	0.207
Birth weight (Kg)*	1.436(0.19)	1.372 (0.16)	1.405 (0.17)	0.425
Out born/inborn [#]	13 :16	7: 17	6: 14	0.409
Vaginal delivery**	23 (79.3%)	21 (87.5%)	16 (80%)	0.707
Maternal complications**	7 (24.1%)	3 (14.2%)	5 (25%)	0.490
Feeding regime- EBM: IPF: Mixed [#]	12:3:14	9:2:13	8:1:11	0.963
Days of life to reach full feeds*	6.10 (1.8)	6.54 (2.6)	6.7 (2.7)	0.659
Duration of hospital stay (days)*	15.06 (9.5)	15.45 (9.6)	12.85 (5.7)	0.574

*Value in mean (SD); # number; **number (%). Maternal complications-gestational hypertension, gestational diabetes, hypothyroidism and heart disease in mother, EBM-expressed breast milk, IPF-infant powder formula, Mixed-EBM+IPF. \$Group I-NG-FT changed every 12 hourly, ## Group II-NG-FT changed every 24 hourly, \$\$ Group III-NG-FT changed every 48 hourly.

Table 3: Results of nasogastric feeding tube culture.

Organism isolated numbers (%)	First NG-FT culture*			Second NG-FT culture**			Third NG-FT culture [#]		
	Group I	Group II	Group III	Group I	Group II	Group III	Group I	Group II	Group III
<i>Klebsiella</i>	2	4	2	2	7	5	6	15	4
<i>Klebsiella</i> + <i>Pseudomonas</i>	0	0	4	1	5	3	3	0	4
<i>Klebsiella</i> + <i>E. Coli</i> + <i>Pseudomonas</i>	0	0	2	0	0	2	0	0	1
<i>E. Coli</i>	1	2	0	1	2	2	1	2	2
<i>E. Coli</i> + <i>Klebsiella</i>	0	1	1	0	2	0	0	0	2
<i>E. Coli</i> + <i>Pseudomonas</i>	0	0	1	0	1	0	0	0	1
<i>Pseudomonas aeruginosa</i>	0	1	1	1	1	2	1	4	2
<i>Acinetobacter</i>	0	1	0	0	1	1	0	2	1
<i>Acinetobacter</i> + <i>Klebsiella</i>	0	0	0	0	1	0	0	1	1
<i>Acinetobacter</i> + <i>Streptococcus</i>	0	0	0	0	1	0	0	0	0
<i>Coagulase negative staphylococci</i>	0	0	3	2	0	2	2	0	2
Total	3 (10.3)	9 (37.5)	14 (70)	7 (24.1)	21 (87)	17 (85)	13 (44)	24 (100)	20 (100)
P value of total organism growth	Group I vs II	0.0432		0.0001			0.0001		
	Group I vs III	0.0001		0.0001			0.0002		
	Group II vs III	0.0649		0.809			1		

*First NG-FT culture after 12, 24 and 48 hours of NG insertion in group I, II and III respectively, ** Second NG-FT culture after 7 days of NG feeding, # Third NG-FT culture after 14 days of NG feeding.

In the first NG-FT cultures the organism growth was 10.3%, 37.5% and 70% in groups I, II and III respectively. Significant increase in growth of microorganisms on NG-FT was seen when frequency of changing NG-FT was reduced from every 12 to 24 or 48

hours (p = 0.0432 and 0.0001 in group I vs II and I vs III respectively).

Culture positivity had significantly increased in all groups in second (24.1%, 87% and 85% in groups I, II

and III respectively) and third NG-FT cultures (44% in group I and 100% in groups II and III) as compared to first culture, but this was significantly high in groups II and III as compared to group I (p = 0.0001) (Table 2). Out of 219 NG-FT culture reports, 128 (58.44%) had organisms growth (single organism growth in 90 samples and polymicrobial growth in 38 samples). *Klebsiella pneumoniae* was the most common organism isolated in 81 (63.28%) samples [as a single organism in 47 (36.7%)

samples and with polymicrobial growth in 34 (26.5%) samples]. It was found to be resistant to most of the antibiotics, except *Polymyxin B*, to which 42 out of 81 were sensitive.

Pseudomonas aeruginosa was 2nd most common organism isolated in 41 (32%) samples [as a single organism in 13 (10.1%) samples and with polymicrobial growth in 28 (21.8%) samples].

Table 3: Antibiotic sensitivity pattern of nasogastric feeding tube culture.

Antibiotic sensitivity	Klebsiella	E. Coli	Pseudomonas aeruginosa	Acinetobacter	Coagulase negative staphylococci
β-Lactam*	6	3	0	2	2
β-Lactam + BLI combination*	13	4	5	5	1
Cephalosporin's*	15	11	6	3	1
Carbapenems*	26	16	9	6	0
Aminoglycosides*	21	4	4	4	0
Sulphonamides	6	1	0	3	0
Polymyxin B	42	19	13	7	0
Fluroquinolones*	4	0	4	0	2
Linezolid	0	0	0	0	10
Aminopeptides*	0	0	0	0	8
Aztreonam	18	0	12	0	0

*β-Lactam-Amoxicillin, β-Lactam + BLI combination-Ampicilline+Sulbactam, Piperacillin/Tazobactam, Cephalosporin's-Cefotaxime, Ceftriaxone, Ceftazidime, Cefepime, Cefixime, Cefaperazone, cefuroxime, cefopodoxime, Carbapenems- Imipenem, Meropenem, Aminoglycosides-Amikacin, Gentamycin, Tobramycin, Fluroquinolones-Ciprofloxacin, Ofloxacin, Aminopeptides-Vancomycin, Teicoplanin

Table 4: Comparison of outcome parameters.

Outcome	Group I (n=29)		Group II (n=24)		Group III (n=20)		P values	
	Positive#	Negative#	Positive#	Negative#	Positive#	Negative#	Positive#	Negative#
Number of episodes of feed intolerance*	1 (0,2)	1 (0,2)	1 (0,2)	1 (0,2)	1 (0,2)	1 (0,2)	1	1
NEC**	4 (19)	1 (12)	1 (5)	2 (50)	3 (21)	1(16)	0.310	0.269
Culture proven Sepsis**	3 (14)	1 (12)	6 (30)	1 (25)	2 (14)	1 (16)	0.374	0.851
Mortality**	2 (9)	0	1 (5)	0	1 (7)	0	0.904	1

NG-FT Culture, *Median, **Numbers (%)

Pseudomonas aeruginosa was also resistant to most of the antibiotics except Polymyxin B and Aztreonam (13 and 12 out of 41 respectively). *E. coli* were isolated from 27 (21%) NG-FT culture reports [as a single organism in 13 (10.1%) samples and with polymicrobial growth in 14 (10.9%) samples]. It was found to be sensitive to *Polymyxin B* (19 out of 27), Carbapenems (16 out of 27) and Cephalosporins (11 out of 27). Coagulase negative Staphylococci (CONS) and Acinetobacter were isolated in 11 and 10 culture reports respectively (Table 3). There was no significant difference in episodes of feed intolerance, NEC, culture proven sepsis and mortality irrespective of NG-FT culture report in all three groups (Table 4).

DISCUSSION

In the present study, the increase in organism growth on NG-FT, with reduced frequency of tube change, can be due to environmental flora of NICU which is transmitted to newborns through health care personnel and mother. It can also be attributed to long stay in NICU as seen in 2nd and 3rd NG-FT cultures. *Klebsiella pneumoniae* (63.28%), the most common organism isolated from NG-FT culture, is also the most common organism responsible for neonatal sepsis at our institute. We found antimicrobial drug resistance to conventional antibiotics among all Gram Negative and Negative isolates.

Hurrell et al found significant difference between frequency of NG-FT change ($p < 0.001$) and organism growth with maximum bacterial counts recorded at 48 hours. As in the present study, *Enterobacteriaceae* were isolated from majority (76%) of samples in his study.¹⁴

Petersen et al found that neither the presence of contamination, nor the density, was associated with the time the NG-FT had been in use. 89% NG-FT culture yielded significant bacterial growth; most common bacteria were *Enterococcus* spp. and CONS. 55% yielded pathogenic bacteria *Enterobacteriaceae* and *Staphylococcus aureus*. The difference in the bacteriological profile of this study from the present study can be explained by the fact that the first culture was sent at 7th day for all cases.¹⁵

Das et al showed that colonization of Gram negative bacilli (GNB) in neonatal gut is driven mainly by external factors as a stay in the NICU (environmental) and prolonged feeding by a tube. Gastric aspirates and stool sample were examined for gut colonization. Within 7 days of birth, GNB (most commonly *Klebsiella Pneumoniae*) were found in the gut of > 99% newborns. This study revealed an association of gut colonization with neonatal sepsis. Difference in result from the present study might be because of inclusion of septic neonates also in their study.⁵

Mehall et al found that 71 of 125 NG-FTs were contaminated and concluded that bacterial contamination of enteral feeding tubes occur frequently, causes significant feeding intolerance and may contribute to NEC.¹⁶

We didn't find any correlation between organism growth and development of feeding intolerance, necrotizing enterocolitis (NEC), neonatal sepsis and mortality in all three groups. This can be due to the reason that we had excluded all septic preterm newborns from present study; only those who required NG-feeding and were vitally stable were included.

Limitation of present study was small samples size. More studies are required to see any correlation between contamination of NG-FT and development of feeding intolerance, NEC and sepsis. This study highlights that frequent changing of NG-FT reduces the incidence of contamination of feeding tube.

Organisms which are present in NICU environment commonly contaminate NG-FT through health care personnel and caregivers. Micro-organism growth on NG-FT is not always associated with increased frequency of feeding intolerance, NEC and sepsis.

CONCLUSION

This study highlights that frequent changing of NG-FT reduces the incidence of contamination of feeding tube.

Organisms which are present in NICU environment commonly contaminate NG-FT through health care personnel and caregivers. Micro-organism growth on NG-FT is not always associated with increased frequency of feeding intolerance, NEC and sepsis.

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Conflict of interest: None declared

Ethical approval: The study was approved by the Institutional Ethics Committee

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