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# To Determine The Frequency Of Necrotizing Enterocolitis In Preterm Infants Treated With Prophylactic Probiotics Versus Controls

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## Abstract

**Objective:** To determine the frequency of necrotising enterocolitis in preterm infants treated with prophylactic probiotics versus controls.

**Methods:** This was a Quasi experminet, conducted in Holy Family Hospital Rawalpindi, Department of Paediatrics from Jan 2020 to Dec 2020. A total of 170 preterm neonates (gestational age 30–35 weeks, birth weight <1500 g) were enrolled and randomly divided into two groups: Group A received prophylactic probiotics, while Group B received routine management without probiotics. Both groups followed standard feeding protocols using expressed breast milk or preterm formula, gradually advancing feeds as tolerated. NEC was monitored clinically and radiologically and confirmed by pneumatosis intestinalis. The study endpoints included NEC incidence with follow-up until 10 days of life or discharge. Data were analysed using SPSS 23, with chi-square tests applied post-stratification for variables such as gestational age, gender, and weight. A p-value of <0.05 was considered statistically significant.

**Results:** Comparison of frequency of necrotising enterocolitis in preterm infants treated with prophylactic probiotics versus controls shows 4.71%(n=4) NEC in Group A and 18.82%(n=16) in Group B.

**Conclusion:** The frequency of necrotizing enterocolitis in preterm infants treated with prophylactic probiotics is lower when compared with controls

**Keywords:** Preterm, probiotics, necrotising enterocolitis

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## Introduction

Necrotizing enterocolitis is one of the common medical emergencies which involve the gastrointestinal tract, primarily affecting premature neonates.[1] In necrotizing enterocolitis there is inflammation and necrosis of the walls of the gastrointestinal tract including small and large intestines with translocation of the gas-forming microorganism from the gastrointestinal tract.[2]

The incidence of necrotizing enterocolitis has an inverse relationship with gestational age and birth weight with an overall incidence of 5 to 7 percent for all infants ≤33 weeks gestational age and weighing less than 1500g at birth. However, the incidence varies from country to country; in Japan, the incidence is lower as compared to Sweden i.e. 16% vs 22% of the very low birth weight.[3]

Necrotizing enterocolitis is one of the most devastating diseases of neonates with high morbidity and mortality. It is estimated that mortality from necrotizing enterocolitis ranges from 15 to 30 per cent. Necrotizing enterocolitis is a multifactorial disease with various risk factors including prematurity, low birth weight, decreased immunity, ischemia/reperfusion injury, abnormal gut bacterial colonization, delay in enteral feeding of human milk and wide use of broad-spectrum antibiotics.[4]

Approximately 90% of infants with necrotizing enterocolitis are born preterm. Those who survive the high mortality of necrotizing enterocolitis are at increased risk of various complications including impaired neurodevelopment, short gut syndrome and strictures.[5]

The signs and symptoms of necrotizing enterocolitis vary, they may be subtle including nonspecific slight distention of the abdomen, vomiting, diarrhoea, irritability with apnea or bradycardia and changes in the appearance/activity of the infant or there may be systemic inflammation, bowel perforation and shock depending on the severity of the disease.[6] The diagnosis of necrotizing enterocolitis is usually made based on signs and symptoms supported by haematological and radiological investigations. The presence of air in the walls of the intestine is a path gnomic of necrotizing enterocolitis.[7]

Different prophylactic treatment modalities have been tried for the prevention of necrotizing enterocolitis in premature newborns which include withholding enteral feeding, use of enteral antibiotics, feeding of mother's expressed breast milk and use of various growth factors and glucocorticoids.[8] Recently use of probiotics has been shown to decrease the incidence of necrotizing enterocolitis.

This study will provide new insights into the efficacy of prophylactic probiotics in reducing necrotizing enterocolitis (NEC) among preterm infants in Pakistan, complementing existing research from other hospitals. Focusing on our specific group of population and employing specific probiotic strains, will add valuable data to the national understanding of NEC prevention strategies to reduce the burden of prematurity-associated complications in neonates.

## Materials And Methods

This was a Qusai experiment which was conducted in Holy Family Hospital Rawalpindi, Department of Paediatrics from Jan 2020 to Dec 2020. The sample size was calculated using the WHO sample size calculator using the following parameters: Level of significance=5%, power of the test=90%, anticipated Population, proportion P1=0.025, anticipated Population, proportion P2=0.151,9 with sample size=85 in each group. A consecutive non-probability method was used for sampling.

Preterm neonates (between 30 to below 36 weeks' gestation) of either gender with birth weight less than 1500g and surviving the first 48 hours of life were included in the study. Gestational age was calculated by using the date of the last menstrual period. Newborns with clinical signs of sepsis, congenital anomalies and birth asphyxia were excluded. The study was started after approval from the ethical committee of our hospital. Informed consent was taken from the parents after explaining in detail the purpose of the study. Patients were divided into two groups randomly based on the lottery method. In group A patient was given prophylactic probiotics. In Group B patients were managed routinely without prophylactic probiotics. A senior nurse in the nursery was requested to administer the probiotic to the babies allocated to group A. Probiotic drops were administered in the form of drops from the ampoule mixed with the feed. Half an ampoule was given in the morning and half in the evening. Probiotics were started once the baby was tolerating feed for 24 hours. Routine management includes keeping the baby's nil per oral for 24 hours after birth, IV fluids and IV antibiotics. Babies who remained well for 24 hours after birth were then started on feed. Feeding protocols were followed strictly in both groups. All the babies were fed on either breast milk or preterm formula milk. Feed began with trophic feeds that were continued for 24 hours and if the baby remained well the feed was gradually built up by an increment of 10 to 20 ml/kg/day till feeding was fully established at 150ml/kg/day. Feed intolerance was defined as nasogastric aspirates of more than half of the amount of the previous feed with or without abdominal distension. Patients were monitored for development of necrotizing enterocolitis. NEC was suspected in babies presenting with a triad of feed intolerance, abdominal distension and blood in stools. Diagnosis of NEC was later confirmed by the presence of air in the wall of the intestine (pneumatosis intestinalis). The patients enrolled in the study were followed up in both groups till 10 days of life or until discharge whichever came first. The person collecting the data (postgraduate trainee in the ward) and parents of enrolled babies were unaware of the group allocation of study subjects ensuring blinding. The data was collected in the form of variables on a predesigned proforma and analyzed using SPSS 23. Frequencies and percentages were calculated for qualitative data like gender and NEC. Mean and standard deviation were calculated for quantitative data i.e. age of gestation and APGAR score, weight. Chi chi-square test was applied to compare the difference between (the frequency of NEC) in the two groups. Effect modifiers like gestational age, gender, and weight were collected by stratification. Post-stratification chi-square test was applied. A p-value $<0.05$ was considered statistically significant.

## Results

A total of 170 preterm infants were included in the study, evenly divided into two groups (85 in each). The gender distribution revealed a nearly equal proportion of males and females in both groups. Group A comprised 51.76% (n=44) males and 48.24% (n=41) females, while Group B included 55.29% (n=47) males and 44.71% (n=38) females.

Gestational age distribution showed that in Group A, 51.76% (n=44) of infants were between 30-32 weeks, and 48.24% (n=41) were between 33-35 weeks. Similarly, Group B had 58.82% (n=50) of infants aged 30-32 weeks and 41.18% (n=35) aged 33-35 weeks. The mean gestational age was 32.87±1.54 weeks in Group A and 32.53±1.57 weeks in Group B. The primary outcome of

the study was the frequency of necrotizing enterocolitis (NEC) in preterm infants. Results demonstrated a significantly lower incidence of NEC in Group A, where 4.71% (n=4) of infants developed NEC, compared to 18.82% (n=16) in Group B. Conversely, 95.29% (n=81) of infants in Group A and 81.18% (n=69) in Group-B showed no signs of NEC, with a p-value of 0.004, indicating a statistically significant difference between the groups. (Table 1)

**Table 1: Comparison of frequency of necrotizing enterocolitis in preterm infants treated with prophylactic probiotics versus controls (n=170)**

| NEC | Group-A (n=85) | | Group B (n=85) | |
|---|---|---|---|---|
| | No. of patients | % | No. of patients | % |
| Yes | 4 | 4.71 | 16 | 18.82 |
| No | 81 | 95.29 | 69 | 81.18 |
| Total | 85 | 100 | 85 | 100 |

*P value: 0.004*

Effect modifiers such as gestational age, gender, and weight were analysed through stratification. A post-stratification chi-square test was applied, and results with a p-value of less than 0.05 were considered statistically significant. The detailed results of these analyses are presented in Tables 2-4.

**Table 2(a): Stratification for with regards to gestational age (n=170)**
**G. AGE: 30-32 weeks**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.04 |
| A | 2 | 42 | |
| B | 9 | 41 | |

**Table 2(b): Stratification for with regards to gestational age (n=170)**
**G. AGE: 33-35 weeks**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.04 |
| A | 2 | 39 | |
| B | 7 | 28 | |

**Table 3(a): Stratification for with regards to gender (n=120)**
**Male**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.03 |
| A | 2 | 42 | |
| B | 9 | 38 | |

**Table 3(b): Stratification for with regards to gender (n=120)**
**Female**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.05 |
| A | 2 | 39 | |
| B | 7 | 31 | |

**Table 4(a): Stratification for with regards to weight (grams) (n=170)**
**Weight: >1000-1500grams**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.25 |
| A | 3 | 41 | |
| B | 7 | 43 | |

**Table 4(b): Stratification for with regards to weight (grams) (n=170)**
**Weight:500-1000 grams**

| Group | NEC | | P value |
|---|---|---|---|
| | Yes | No | 0.002 |
| A | 1 | 40 | |
| B | 9 | 26 | |

## Discussion

The study evaluated the impact of prophylactic probiotics on the incidence of necrotizing enterocolitis (NEC) among preterm infants, comparing outcomes across groups with similar distributions of gender and gestational age (30–35 weeks). The findings demonstrated a significantly lower incidence of NEC in the probiotics group compared to the control group, corroborating evidence from both national and international studies over the past five years that highlight probiotics as an effective intervention to improve neonatal outcomes.

A local study conducted at Abbasi Shaheed Hospital in Karachi, Pakistan, found that probiotics reduced NEC incidence to 4% compared to 16% in the control group. This reduction was accompanied by declines in mortality and sepsis rates, as well as improved growth and shorter hospital stays in the probiotics group. The study further noted that gestational age per week and birth weight per 100 grams had a significant effect on NEC incidence, with p-values of 0.028 and 0.018, respectively.[9]

Another study from Bahawalpur, Pakistan, demonstrated similar results, with NEC occurring in 6.52% of neonates in the probiotics group versus 23.9% in the placebo group (p=0.020). NEC in the probiotics group was significantly associated with gestational age of 31–32 weeks (p=0.012), spontaneous vaginal delivery (p=0.028), and higher maternal education (p=0.006). In the placebo group, NEC was more common among neonates with a birth weight of 1–1.5 kg (p=0.007). These findings highlight differences in risk factors between probiotics and placebo groups, which may vary based on demographic and clinical contexts.[10]

While our study did not examine maternal factors like education or delivery methods, we identified male gender and birth weight below 1000 grams as significant risk factors for NEC, with p-values of 0.03 and 0.002, respectively. Gestational age ranges of 30–32 weeks and 33–35 weeks had significant, though similar, effects on NEC incidence in both groups (p=0.04). These results align with findings from local studies, emphasizing the protective effect of probiotics in reducing NEC.

Chowdhury et al. added further evidence by showing that probiotics significantly reduced NEC incidence among very low birth weight (VLBW) infants, alongside faster achievement of full oral feeding and shorter hospital stays. These findings highlight the broader benefits of probiotics in reducing healthcare burdens while improving neonatal health outcomes.[11]

Internationally, a 2019 meta-analysis of randomized controlled trials revealed that probiotics reduce severe NEC and all-cause mortality among preterm infants. This meta-analysis also pointed out variability in probiotic strains, dosages, and treatment durations across studies, suggesting the need for standardized protocols.[12]

A 2024 review in Current Treatment Options in Pediatrics expanded on these findings, noting reductions in late-onset sepsis and mortality, alongside NEC. While our study focused on NEC prevention, these broader findings suggest a multifaceted role for probiotics in neonatal care.[13]

One limitation of our study is the narrow scope, as it did not explore secondary benefits such as reduced mortality, sepsis rates, or hospital stays, which were highlighted in other studies. Future research in Pakistan should address these gaps, with an emphasis on optimizing probiotic strains, dosages, and treatment durations.

In summary, our study reinforces the efficacy of probiotics in reducing NEC and aligns with both local and international findings.[15]

## Conclusions

We concluded that the frequency of necrotizing enterocolitis in preterm infants treated with prophylactic probiotics is lower when compared with controls.

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