ORIGINAL ARTICLE

Effect of oral colostrum application every 2 hours and 4 hours in order to achieve trophic feeding in preterm infants: A randomized controlled trial

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ABSTRACT

Introduction: Enteral feeding in preterm neonates starts with trophic feeding, which is the practice of feeding minute volumes of enteral feeds (starting at 10-25mL/kg/day) through an orogastric tube. Colostrum has protective effects, such as anti-inflammatory, immunomodulatory, and antimicrobial effects. The oral colostrum application is a safe, effective and economical therapy. However, the most optimal frequency of the oral colostrum application is not yet conclusive. This study aims to evaluate the effects of applying colostrum orally every 4 and 2 hours in order to achieve trophic feeding in preterm infants.

Materials and Methods: In this randomized controlled trial with an open-label design, very-low-birth-weight neonates admitted to RSUP Dr. Sardjito from March to August 2023 were allocated to receive oral colostrum applications either every two hours or every four hours. Subjects were randomized into study groups using a random block size of four through computer-generated in a 1:1 ratio. The primary outcome was the time to achieve trophic feeding. The extraneous variables were necrotizing enterocolitis, sepsis, hemodynamically significant Patent Ductus Arteriosus (hsPDA) and gender. Data analysis was conducted using SPSS.

Results: A total of 40 neonates were analyzed for primary outcome. Of these, 20 neonates received oral colostrum applications every 2 hours, and the other 20 subjects were fed every 4 hours. Bivariate analysis showed that colostrum application given every 4 hours achieved the trophic feeding 0.47 day faster than the colostrum application every 2 hours. However, the difference between the two feeding methods was not statistically significant (p=0.703).

Conclusion: There is no significant difference in achieving trophic feeding in preterm neonates (less than 34 weeks) whether the colostrum was given every 2 or 4 hours.

KEYWORDS:
Colostrum, very low birth weight, trophic feeding, clinical trial, preterm

INTRODUCTION

A healthy oral cavity serves as the first line of defense against infections. When enteral feeding via oral cannot be administered to the sick or preterm infants, it may compromise the integrity of the buccal mucosa and serve as a focal point for infection. The oropharyngeal colostrum application helps maintain oral cavity moisture and reduces bacterial colonization.[1]

Numerous studies have evaluated the effects of the oral care using sterile water, colostrum, or breast milk. The usage of colostrum or breast milk is preferred due to its immunological benefits. The oropharyngeal colostrum application with breast milk involves the direct administration or spreading of breast milk onto the oral mucosa to keep it clean and intact. This procedure is safe and well-tolerated, even in clinically unstable infants.[1,2]

Applying colostrum to the oral cavity stimulates the immune response of the oropharyngeal lymphoid tissue and prevents microbial adhesion by providing a layer of immunoglobulin A (IgA) on the oropharyngeal mucosa. Oropharyngeal colostrum immunotherapy is a good strategy to deliver antimicrobial and anti-inflammatory protective factors, stimulate immune cells in oropharyngeal lymphoid tissue, support immune system maturation, and aid digestive system maturation.[3,4] Additionally, it is beneficial for the development of taste and smell sensations, initiates early enteral feeding, accelerating full enteral feeding, and speeds up the return to birth weight.[1,2]

The application of colostrum to the oropharynx is a simple, economical, and positive treatment, but the frequency of colostrum application is still inconclusive. Further studies are needed for producing reliable scientific evidence. Some studies indicate that more frequent colostrum application in the oropharynx leads to better outcomes.[5] A meta-analysis in June 2022 by Huo et al., including 11 randomized controlled trials, showed that oropharyngeal colostrum administration reduces the risk of necrotizing enterocolitis, late-onset sepsis, ventilator-associated pneumonia, accelerates full enteral feeding, and reduces the length of hospital stay.[6] A more recent meta-analysis in July 2022 by Cai et al. stated similar findings.[7] In both, conducted meta-analyses, variations in the

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interval of oropharyngeal colostrum administration were observed among studies.[6,7]

Most oropharyngeal colostrum application is done at intervals of every 2 hours. However, there is another study using a 4-hour interval, proving that colostrum administration every 4 hours increases lactoferrin in saliva and has a positive impact on expediting enteral feeding.[4] Hence, oral colostrum application has positive effects, but currently there is no research on the optimal frequency of colostrum application that can be applied in Indonesia.

This study aims to evaluate the effects of oral colostrum application frequency, every 4 hours and every 2 hours, in order to achieve trophic feeding in preterm infants <34 weeks gestational age.

MATERIALS AND METHODS

This experimental study using an open-label Randomized Controlled Trial (RCT) design was conducted from March to August 2023. The study was done in a level III neonatal referral center in Indonesia at Neonatal Intensive Care Unit (NICU) of RSUP Dr. Sardjito. Inclusion criteria were infants with gestational age <34 weeks, birth weight between 1000-1500 grams, colostrum used was from the mother's own breast milk (not donors' milk), and parents/guardians willing to participate in the study by signing informed consent. This study excluded infants with major congenital abnormalities (Table I), infants born to Human Immunodeficiency Virus-positive mothers, infants whose mothers consumed breastfeeding contraindicated drugs (Table I), severely ill mothers unable to provide breast milk, and mothers unable to supply breast milk within 48 hours after delivery. The research was conducted after obtaining approval from the Ethics Committee of the Faculty of Medicine, Public Health, and Nursing, UGM, with approval number KE-FK-0811-EC-202. The trial was registered retrospectively at the U.S. National Institutes of Health (ClinicalTrials.gov) with number NCT06379178.

In this study, data was collected based on consecutive sampling until the required sample size was reached. The sample size was determined using the formula for the independent t-test with alpha= 0.05 and beta=0.2, we calculated a minimum of 14 samples in each group, totaling 28.[8] To account for potential loss to follow-up, the researcher added 10% more samples, resulting in a minimum total of 32 samples. Subjects were randomized into study groups using a random block size of four through computer-generated in a 1:1 ratio. This study used four randomization blocks, with code A for colostrum application every 2 hours and code B for colostrum application every 4 hours. There were six permutation sequences: AABB, ABBA, BAAB, BABA, ABAB, BBAA. Samples were subsequently allocated to either the 2-hour or 4-hour colostrum application group based on the code in the randomized sequence. This study was also an open-label controlled trial with no blinding.

The study necessitated colostrum from the mother's own breast milk, a 1 ml syringe to measure colostrum volume, a tool to facilitate even colostrum application, and a suction catheter to remove excess saliva. Breast milk expression was initiated within 6 hours post-delivery, adjusted based on the mother's condition. Nurses or doctors assisted with the initial breast milk expression using standard hospital-grade breast pumps or manual methods. Expression was performed by the mother or with the assistance of her spouse every 2-3 hours (eight times per day), with a minimum duration of 10 minutes on each breast.

The expressed breast milk was collected in a container, drawn into a 1 ml, 3 ml, or 5 ml syringe depending on the obtained volume, and then labeled for identification. For colostrum application, the nurse took 0.2 ml of colostrum using a 1 ml syringe. Colostrum application prioritized the freshest breast milk, allowing direct administration to the research subject. The remaining breast milk can be used within 2-4 hours at room temperature (16-29 °C) or stored in the refrigerator or freezer according to breast milk storage standards. If frozen, the breast milk should be thawed in the refrigerator before being extracted in the amount of 0.2 ml using a 1 ml syringe for application to the research subject. If stored in the refrigerator, rather than the freezer, 0.2 ml of breast milk should be drawn with a 1 ml syringe, allowed to sit for a designated period (15-30 minutes), and subsequently administered to the research subject.[9]

The nurses followed a pre-procedure checklist for colostrum application in the NICU, prepared and administered the colostrum for each research subject scheduled for colostrum application. If the research subject used mechanical ventilation, the colostrum application procedure was carried out by two nurses. Before colostrum application, the oral cavity of the research subject was examined. If excess fluid or saliva was found, suction was performed first, or a sterile gauze was used to remove excess fluid while eliminating dry skin or debris on the lips. Then, with the aid of a 1 ml syringe after removing the needle, colostrum was evenly applied to both right and left cheeks (buccal mucosa), the roof of the mouth, gingival surface, and lips for approximately 1-2 minutes every 2 hours (12 times per day) or every 4 hours (6 times per day) depending on the code in the randomization sequence. Colostrum in the mouth will be distributed as it mixes with saliva. Colostrum application to the oral cavity began within 24-48 hours after birth and continued for a total of 5 days.

In case of an unstable clinical condition such as apnea, tachypnea, or desaturation, stabilization was carried out first following the Neonatal Intensive Care Unit (NICU) RSUP Dr. Sardjito Standard Operational Procedure (SOP). After stabilization, colostrum application can be resumed. In cases of feeding intolerance, management was carried out according to the SOP for handling feeding intolerance. If breast milk is available and there are no contraindications to enteral feeding, it can be administered promptly.

Observations of subjects were recorded using the NICU daily monitoring sheet, with detailed notes on various evaluations such as age, days of care, vital signs, feeding intolerance, diagnosis, fluid balance, diuresis, and so forth. Primary research data was from the Case Report Form (CRF) based on the Electronic Medical Record (EMR) and NICU daily

Table I: Exclusion criterias

Major congenital abnormalities			
Atresia esophagus	Atresia anorectal	Association Atresia intestinal	Anencephaly
Conjoined twins	Cyanotic congenital heart disease	Diaphragmatic hernia	Gastroschisis
Hypoplasia Lung	Klinefelter	Lung agenesis	Omphalocele
Orofacial cleft	Pyloric stenosis	Syndrome Hirschrpung disease	Trisomy 13
Trisomy 18	Turner syndrome		
Contraindicated drugs			
Abacavir	Atenolol	Simvastatin	Valganciclovir
Acitretin	Azathioprine	Dihydroergotamine	Macitentan
Alprazolam	Ado-trastuzumab emantasine	Diphenoxylate	Methamphetamine
Amitriptyline	Amantadine	Dolutegravir	Miltefosine
Amoxapine	Amphetamine	Efavirenz	Nelfinavir
Aripiprazel	Anakinra	Emtricitabine	Nevirapine
Aspirin	Atazanavir	Enfuvirtide	Pentamidine
THC (marijuana)	Atorvastatin	Entecavir	Rifabutin
Tenofovir	Chlorpromazine	Ergotamine	Ritonavir
Tramadol	Clonazepam	Fosamprenavir	Saquinavir
Lopinavir	Cobicistat	Heroin	Smallpox vaccine
Lamivudine	Cocaine	Indinavir	
Zidovudine	Delavirdine	Stavudine	

Table II: Baseline Characteristics of Research Subjects

Characteristic	Colostrum application every 2 hours	Colostrum application every 4 hours
Gender, n (%)		
Female	6 (15)	11 (27,5)
Male	14 (35)	9 (22,5)
Birth weight in g, mean ±SD	1.314,80 ± 117,921	1.215,30 ± 140,067
Gestational age in weeks, n (%)		
28 weeks - <31+6 weeks	16 (40)	10 (25)
32 weeks - <33+6 weeks	4 (10)	10 (25)
Methods of delivery, n (%)		
Spontaneous delivery	11 (27,5)	6 (15)
Abdominal delivery	9 (22,5)	14 (35)
Maternal infection risk factors, n (%)		
With risk factors	14 (35)	12 (30)
Without risk factors	6 (15)	8 (20)
Maternal age in years, mean ±SD	31,45± 6,022	32,15± 7,013
Sepsis, n (%)		
Yes	20 (50)	19 (47,5)
No	0 (0)	1 (2,5)
hemodynamically significant Patent Ductus Arteriosus, n (%)		
Yes	5 (12,5)	6 (15)
No	15 (37,5)	14 (35)
Necrotizing enterocolitis, n (%)		
Yes	3 (7,5)	1 (2,5)
No	17 (42,5)	19 (47,5)

g=gram; SD= standard deviation

Table III: Bivariate analysis (Independent t-test) of colostrum application and days to achieve trophic feeding

Variable	Mean (days) to achieve trophic feeding		95% CI	p-value
	2 hours	4 hours		
Colostrum application	7	6,53	-2,011 -2,944	0,703

monitoring sheet. The research subjects were prospectively observed, with data being regularly monitored every day until the primary outcomes were achieved, with a maximum observation period of 21 days.

The primary outcome is the time to achieve trophic feeding defined as reaching a target volume of 25mL/kg/day with enteral feeding tolerance. Tolerance of enteral feeding was assessed if gastric residual was ≤5mL/kg or residual was <50% of the volume given, no blood was found in the residual, no vomiting or abdominal distension occurred, and there was no decrease, delay, or cessation of enteral feeding. While external variables include necrotizing enterocolitis, sepsis, hsPDA, and gender, represented as categorical variables.

Table IV: Bivariate analysis (Independent t-test) of external variable and dependent variable (days to achieve trophic feeding)

Variable	Mean (days) to achieve trophic feeding		95% CI	p-value
	Yes	No		
Subjects with colostrum application every 2 hours				
Necrotizing enterocolitis	10,50	6,53	-2,292 – 10,225	0,197
Sepsis	7,00	-	-	-
hsPDA	10,33	6,29	-1,093 – 9,188	0,114
Male gender	6,42	8,40	-2,575 – 6,542	0, 368
Subjects with colostrum application every 4 hours				
Necrotizing enterocolitis	9,00	6,36	-3,215 – 8,501	0,348
Sepsis	6,71	4,00	-3,132 – 8,561	0,334
hsPDA	6,00	6,67	-4,430 – 3,096	0,708
Male gender	6,71	6,38	-3,367 – 2,688	0,812

The collected data was entered into a database and processed using Statistical Package for the Social Sciences (SPSS) software version 27. Data characteristics were analyzed using univariate tests to determine frequency distribution. Hypothesis testing involves bivariate analysis to determine the mean time to achieve trophic feeding (Independent t-test). A significance level of $p<0.05$ is considered statistically significant. The study followed the principle of intention to treat, meaning that the entire sample will be analyzed.

RESULTS

There were 47 preterm infants <34 weeks gestational age being treated in the Neonatal Intensive Care Unit (NICU) at RSUP Dr. Sardjito over a six month period between March and August 2023, including those born at RSUP Sardjito and in external healthcare facilities referred to our unit.

Out of the 47 very low birth weight (VLBW) infants, 7 were excluded, including 1 infant with esophageal atresia, 1 infant born to a critically ill mother who passed away, resulting in no available breast milk, 1 with mother consuming medication contraindicated for breastfeeding, and 4 infants receiving donor breast milk. The 40 eligible VLBW infants were randomly divided into two treatment groups: 20 received colostrum application every 2 hours, while the rest received it every 4 hours based on the randomization sequence from the permutation code. During the follow-up process, 8 subjects died before reaching trophic feeding, with a breakdown of 7.5% mortality in the colostrum application every 2 hours group and 12.5% in the every 4 hours group. The study applied the intention-to-treat principle, so the analyzed sample size included all 40 research subjects. Figure 1 presents the flowchart of the study. Table II shows the baseline characteristics of the neonates.

Table III demonstrates the bivariate analysis result using an Independent t-test of the oral colostrum application and the time to achieve trophic feeding. The analysis results indicate that subjects with colostrum application every 4 hours reached trophic feeding, on average, 0.47 days earlier than subjects with oral colostrum application every 2 hours. However, the difference in means is not statistically significant ($p=0.703$).

Table IV shows bivariate analysis results (Independent t-test) of the external variable with time to achieve trophic feeding. It indicates that subjects with necrotizing enterocolitis, sepsis, male gender, and hsPDA did not show statistically significant differences on the mean to achieve trophic feeding, as evidenced by the $p\geq0.05$ values in both groups of subjects receiving colostrum every 2 hours and every 4 hours (Table IV).

DISCUSSION

Our study was conducted on a population of preterm infants <34 weeks gestation and birth weight less than 1500 grams (VLBW) with the frequency of oral colostrum application every 2 hours and every 4 hours. In previous research, three studies provided colostrum every 2 hours, with two of them were conducted on VLBW populations and one study on extremely low birth weight infants (ELBW). There were six studies administering colostrum every 3 hours, with two of them were conducted on VLBW populations, and four other studies did not mention the weight of the subjects. Two studies administered colostrum every 4 hours, with one study was conducted on ELBW populations and one study did not specify the weight of the subjects.[10] The baseline characteristic for birth weight between the two groups in our study was normally distributed.

The gestational age in our study was 28-31+6 weeks in 65.6% of the cases and 32 weeks - <33+6 weeks in 34.4%, whereas in previous studies, both the researches with colostrum application every 2 hours and 4 hours, had populations with an average gestational age of <34 weeks, with only one study by Rodriguez using a population <28 weeks.[11] Thus, all participants included were within the range of preterm infants studied in previous research.

The colostrum volume used in previous studies was 0.2 ml, with only one study by Romero using a colostrum volume of 0.3 ml.[10,12] In this study, the volume given was 0.2 ml for colostrum administration every 2 hours and 4 hours. The duration of colostrum administration in previous studies varied between 1-3 days, 4-7 days, 8-10 days, and more than 10 days.[10] In this study, the duration of colostrum administration was 5 days for both every 2 hours and 4 hours colostrum applications. To date, the researchers have not found a similar study.

The objective of our current study is to investigate the effects of colostrum application every 2 hours and every 4 hours on the attainment of trophic feeding. We have not found a similar previous study, but there are several studies that have been conducted related to feeding status, including full enteral feeding and feeding intolerance.[4,12,13]

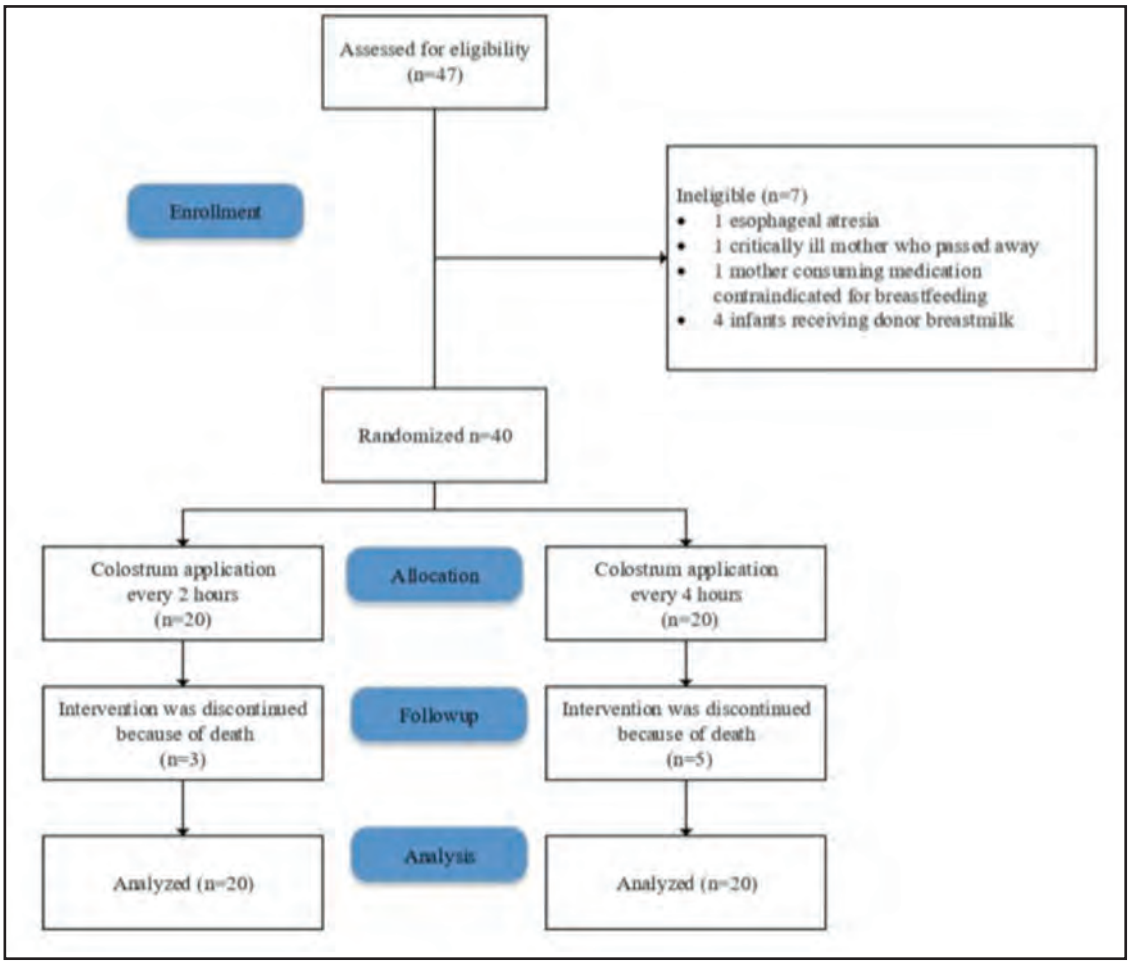

Fig. 1: Research flow until data analysis

The result of a meta-analysis conducted by Fu et al indicates that there is no significant difference in feeding status indicators between colostrum administration every 2 hours and 3 hours.[10] However, when colostrum is administered every 4 hours, a significant difference is observed, with a faster attainment of trophic feeding. These meta-analysis results differ from our study's findings, where the application of colostrum every 2 hours and every 4 hours shows no statistically significant difference (Table III). This discrepancy may be attributed to the differences in the study population, the volume of colostrum used, and the duration of colostrum administration.

The application of colostrum every 2 hours and every 4 hours in the duration of 5 days in patients with necrotizing enterocolitis and sepsis showed no difference in achieving trophic feeding (Table IV). A meta-analysis by Fu et al. suggests that the colostrum application every 4 hours with a shorter duration will affect feeding status, while a duration of 8-10 days can prevent necrotizing enterocolitis and late-onset sepsis.[10] It is possible that our study may show differences if the duration of colostrum administration is longer. Further research is needed to examine the feeding status, necrotizing enterocolitis, or sepsis with colostrum application every 2 hours and 4 hours with a longer duration.

The research results indicate that colostrum application every 2 hours and 4 hours showed no significant difference in achieving trophic feeding. Since our findings show no significant difference in clinical outcome, we suggest that colostrum be administered every 4 hours based on practical considerations, including the limited colostrum availability in the early postpartum. A study conducted in Indonesia examined the average amount of breast milk produced in spontaneous deliveries, with an average of (± 0.155) ml at 2 hours postpartum, (± 1.272) ml at 16 hours postpartum, and (± 1.369) ml at 24 hours postpartum.[14] Another reason for recommending colostrum application every 4 hours is that premature or very low birth weight infants are often born to ill mothers. While NICU workload is not a clinical consideration, the implementation of colostrum application every 4 hours can offer operational benefits, reducing the nursing workload, as providing nutrition is a time-consuming task.[15]

A study indicates that the recommended clinical management for infants is to adopt a minimal handling approach. Minimal handling is a common practice in the NICU to provide restful sleep for infants without disturbance from medical, care, and other examination activities. Minimal handling can prevent the occurrence of intraventricular hemorrhage (IVH) in the acute phase, within

72 hours after birth.[16] The clinical implications of this research could serve as a basis or guideline for clinical practice in colostrum application for very low birth weight infants (VLBW) and suggest that colostrum application should be performed every 4 hours. This aligns with the recommendations from the meta-analysis conducted by Fu et al.[10]

Challenges encountered during our research included difficulties in educating and convincing mothers and families to perform breast milk expression. Often, mothers lacked confidence, so we addressed this issue by providing education to mothers and families immediately after the baby's birth. We also provided intensive family support several times until the 5th day, regularly reminding mothers and families about breast milk expression and collection. On average, after the 4th day, mothers and families became more enthusiastic and disciplined in collecting breast milk.

To the best of our knowledge, this study is one of the first to explore the frequency of oral colostrum application every 2 hours and 4 hours in infants born before 34 weeks of gestation. As of now, we have not found any publications or ongoing studies on the frequency of colostrum applications. However, we acknowledge that our study has some limitations. It was conducted at a single center and had an abnormal distribution of data. The research method used was the randomized control trial open label, and blinding was not possible due to the limitations in the research location and team. While the absence of blinding increases the potential for a placebo effect, we minimized the bias by using objective outcome measures and implementing a standardized procedure checklist. Karyotyping was not performed to determine major congenital anomalies in subjects meeting the exclusion criteria due to the limitations in diagnostic facilities. Subjects meeting the exclusion criteria with a list of major congenital anomalies were determined based on clinical manifestations of related syndromes.

CONCLUSION

To conclude, the frequency of oral colostrum application every 4 hours does not differ from every 2 hours in order to achieve trophic feeding in infants born before 34 weeks of gestation. Colostrum application in the oral cavity should preferably be done every 4 hours because the amount of breast milk produced in the early postpartum period is not abundant. Premature or very low birth weight infants are often born to ill mothers. The implementation of colostrum application every 4 hours is a minimal handling approach compared to every 2 hours and can reduce the workload of NICU nurses. Further research is needed to evaluate the impact of longer oral colostrum administration durations on feeding status, necrotizing enterocolitis, or late-onset sepsis in colostrum applications every 2 hours and 4 hours.

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SCOPE STATEMENT

This original research presents a critical aspect of neonatal care — evaluating the effects of oral colostrum application frequency, administered every 4 hours versus every 2 hours, on achieving trophic feeding in preterm infants <34 weeks gestational age by using an open-label randomized clinical trial. The study explored numerous factors, including gestational age, birthweight, gender, maternal age and risk factors, methods of delivery, necrotizing enterocolitis, sepsis, hemodynamically significant patent ductus arteriosus among the subjects. Providing evidence into the most effective frequency of colostrum administration for these vulnerable infants, the study highlights the potential benefits for implications in clinical practice in optimizing feeding status of preterm infants. The study findings hold a promising suggestion in protocols and practices within neonatal intensive care settings.

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