The Effect of Maternal Milk on Neonatal Morbidity of Very Low-Birth-Weight Infants

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Objective: To examine the dose effect of maternal milk on neonatal morbidity of very low-birth-weight (<1.5 kg) infants.

Design: Prospective observational study.

Setting: An urban tertiary care neonatal intensive care unit and follow-up clinic.

Population: One hundred nineteen singleton very low-birth-weight infants admitted from January 1, 1997, to February 14, 1999 (mean birth weight, 1056 g; mean gestational age, 28 weeks; 57% male; and 43% white).

Methods: A comparison of the effect on neonatal outcomes of daily graded doses (1-24, 25-49, and ≥50 mL/kg of body weight) of maternal milk through week 4 of life vs a reference group receiving no maternal milk.

Main Outcome Measures: Neonatal outcomes examined included rates of sepsis after age 5 days, retinopathy of prematurity, chronic lung disease, necrotizing enterocolitis, jaundice, duration of ventilator dependence, and length of hospital stay.

Results: Seventy-nine infants (66%) received maternal milk, of whom 32 received at least 50 mL/kg per day through week 4 of life. Poisson regression analysis adjusting for birth weight, sex, and ethnicity revealed that the mean number of episodes of sepsis for infants receiving at least 50 mL/kg per day was lower by a factor of 0.27 (95% confidence interval, 0.08-0.95) compared with infants receiving no maternal milk. There was no effect of maternal milk on other neonatal outcomes.

Conclusions: A daily threshold amount of at least 50 mL/kg of maternal milk through week 4 of life is needed to decrease the rate of sepsis in very low-birth-weight infants, but maternal milk does not affect other neonatal morbidities.
less than 33 weeks, and absence of positive drug screen, major congenital anomaly, intratertiary infection, or insurmountable maternal social factors (including custody dispute, adoption pending, mother visiting from overseas, or mother in shelter because of domestic violence). Of the 149 eligible infants, mothers of 119 (80%) agreed to participate in the study. The participating infants did not differ from the nonparticipating infants with respect to birth weight, gestational age, ethnicity, or sex. This study was approved by the Institutional Review Board of University Hospitals of Cleveland.

Per hospital policy, all mothers were strongly encouraged to provide breast milk for their premature infants. The nutritional protocol in the Neonatal Intensive Care Unit included intravenous fluids with dextrose during the first 24 hours. Enteral intake was begun by day 2 or 3 of life, depending on the severity of the infant’s illness. Intravenous nutrition was continued until a daily enteral intake of 120 mL/kg of body weight was reached. Infants received their mother’s milk in the sequence it was expressed, except that fresh rather than frozen milk was given if available. Maternal milk was fortified, and 100 J/oz of preterm infant formula was offered when the infant reached a daily oral intake of at least 110 mL/kg. Limited availability of maternal milk was the sole reason infants were fed preterm formula in addition to maternal milk. Full feeding was defined as full enteral feeding with no intravenous intake.

DATA COLLECTION

Nutritional intake was recorded daily and included route, volume, and type of intake (intravenous vs oral) and day of first and full enteral feeds. Oral intake included maternal milk, preterm formula, and sterile water given by gastric tube or by mouth. Powdered fortifiers (eg, Enfamil Human Milk Fortifier; Mead Johnson Nutritional, Evansville, Ind) that are added to a given volume of maternal milk were not considered to change that volume. Liquid fortifiers (eg, Similac Natural Care; Ross Products Division, Abbott Laboratories, Columbus, Ohio) that are mixed 1:1 with maternal milk increase the apparent volume of maternal milk received. Therefore, the liquid fortifier was counted as preterm formula, and only the actual volume of maternal milk received was counted as maternal milk. The proportion of total nutritional volume received as intravenous intake, maternal milk, and preterm formula, ranging from 0% to 100% per day for each infant, resulted in a sum total of 100% of the infant’s daily nutritional intake.

Demographic, birth, and neonatal data and information on oxygen and ventilator dependence, postnatal dexamethasone sodium phosphate use and dosage, and neonatal complications, including sepsis, jaundice, necrotizing enterocolitis, and cranial ultrasound abnormalities, were recorded prospectively.

We compared the effect of varying dosages of maternal milk on neonatal outcomes. Classifications of maternal milk intakes included a daily mean of 1 to 24, 25 to 49, and at least 50 mL/kg through week 4 of life, compared with a reference group receiving no maternal milk. Neonatal outcomes examined included the rates of sepsis, necrotizing enterocolitis, chronic lung disease, jaundice (indirect bilirubin, >10 mg/dL), and retinopathy of prematurity, in addition to the mean duration of ventilator dependence and length of hospital stay. To clarify the timing of a potential effect of feeding maternal milk, we also examined the effect of receiving varying amounts of maternal milk through weeks 2, 4, and 6 of life on the rate of sepsis occurring after these time points.

Sepsis was defined as a positive blood culture obtained in the presence of clinical signs or symptoms of infection, treated for 5 or more days with antibiotics. Episodes of sepsis occurring before the fifth day of life were excluded, because most infants received only minimal enteral feeds before this age. We examined rates of sepsis, rather than other categories of infection, because the definition of sepsis is widely agreed on and it is a common event. Criteria for the diagnosis of pneumonia in ventilated and oxygen-dependent preterm infants vary, meningitis is a rare event, and the diagnosis of urinary tract infection depends on the frequency and adequacy of surveillance urine cultures, making these infections less amenable to study. Retinopathy of prematurity was graded by an experienced ophthalmologist according to international classification. Necrotizing enterocolitis was defined according to modified Bell criteria (2 stage II). Chronic lung disease was defined as oxygen dependence at 36 weeks’ corrected age.

STATISTICAL ANALYSIS

We used the t test to compare continuous measures and the χ2 or Fisher exact test to compare categorical data. Poisson regression was used to analyze the effect of varying dosages of maternal milk on the number of sepsis episodes. The relative risk produced by Poisson regression analysis is the ratio of the mean number of episodes of sepsis for infants exposed to varying amounts of maternal milk, relative to no maternal milk. Logistic regression was used to analyze the effect of varying dosages of maternal milk on the rate of other neonatal morbidities. Results are expressed as odds ratios with 95% confidence intervals. Data that were not normal in distribution were log transformed before analysis, when necessary. Results of linear regression analysis for these outcomes (logarithm of duration of ventilator dependence and of length of hospital stay) are expressed as the mean difference in log days, with 95% confidence intervals for infants receiving varying amounts of maternal milk, compared with no maternal milk. For these outcomes, we added 1 before taking logarithms, to avoid taking logarithms of 0. The results of each regression analysis were adjusted for variables known to affect neonatal morbidity and mortality, including birth weight, ethnicity, and sex.

Maternal descriptors and birth data for the 119 study infants have been reported previously. Of the 119 mothers, 114 (96%) delivered at a perinatal center, of whom 29 (24%) were transported antenatally, 96 (81%) received antenatal steroid therapy, 28 (24%) had fetal distress, and 60 (50%) were delivered by cesarean section.

Table 1 presents birth data relevant to the present study. Seventy-nine (66%) of the 119 infants received maternal milk, all of whom also received preterm formula as needed to achieve a full enteral intake. Daily through week 4 of life, 40 infants (34%) received preterm formula only, 29 (24%) received a mean of 1 to 24 mL/kg of maternal milk, 18 (15%) received 25 to 49 mL/kg, and 32 (27%) received at least 50 mL/kg.

Table 2 gives the total volume for all study infants of maternal milk received per infant, the mean daily volume, and the mean proportion of maternal milk of total intake (oral plus intravenous) and of oral intake. The mean time to achieve 418 J/kg per day was 21 days (range, 8-91 days) and to achieve full oral feeds was 28 days (range, 6-123 days). Birth weight was inversely correlated with time to full oral feeds (r = −.56, P < .001), and the longer it took for infants to achieve full oral feeds, the lower the mean volume (in milliliters per kilogram of body weight.
per day) of maternal milk received through weeks 2, 4, and 6 (P < .001 for all). Infants with higher birth weights received larger volumes of maternal milk throughout the study.

Thirty-four infants (29%) had 1 or more episodes of sepsis after day 5 of life. Twenty-six infants (22%) had 1 episode, 6 (5%) had 2 episodes, and 2 (2%) had 3 episodes. The timing of these episodes is shown in the Figure. Of the 44 episodes of sepsis, 32 were due to coagulase-negative staphylococci, 5 to Candida species, 3 to Staphylococcus aureus, 2 to Escherichia coli, 1 to Serratia species, and 1 to group B streptococci.

Table 3 presents rates of neonatal morbidity according to the mean daily volumes of maternal milk received through week 4 of life. Poisson regression analysis adjusting for birth weight, sex, and ethnicity revealed that the mean number of episodes of sepsis for infants receiving at least 50 mL/kg daily was lower by a factor of 0.27 (95% confidence interval, 0.08-0.95) compared with infants receiving no maternal milk. There was no difference in the mean number of episodes of sepsis among infants receiving lesser amounts (1-24 or 25-49 mL/kg) of maternal milk daily compared with infants receiving no maternal milk. The rates of necrotizing enterocolitis, retinopathy of prematurity, chronic lung disease, and jaundice did not differ according to the amounts of maternal milk received. For duration of ventilator dependence and length of hospital stay, we also adjusted for sepsis because sepsis has been shown to directly affect these outcomes. There was no effect of maternal milk on duration of ventilator dependence or on length of hospital stay, after adjusting for birth weight, ethnicity, sex, and sepsis in a logistic regression analysis.

Table 4 presents the rates of sepsis occurring after weeks 2, 4, and 6 of life according to the feeding of varying amounts of maternal milk (daily mean, 0, 1-24, 25-49, or ≥50 mL/kg) before these time points. Birth weight was not adjusted for in the analysis of these rates. The lowest rates of sepsis occurred for infants receiving 25 to 49 mL/kg per day and at least 50 mL/kg per day of maternal milk, with only 1 infant who received at least 50 mL/kg per day developing sepsis. In addition, in a logistic regression analysis that adjusted for birth weight, we found that the greater the mean volume of maternal milk intake (in milliliters per kilogram of body weight per day), the lower the rate of sepsis after weeks 2, 4, or 6 of life developed sepsis after these ages. In comparison, infants who received less than 50% of their total intake as maternal milk by weeks 2, 4, and 6...
In this study, we examined the dose effect of maternal milk on neonatal morbidity among VLBW infants. Infants who had received at least 50 mL/kg per day of maternal milk had a significantly lower rate of sepsis than infants who had received no maternal milk. There was no reduction in sepsis among infants receiving lesser amounts of maternal milk. We found no effect of maternal milk on length of hospital stay or other neonatal morbidities, including necrotizing enterocolitis, retinopathy of prematurity, jaundice, chronic lung disease, and duration of ventilator dependence. The mean daily volume of maternal milk received through weeks 2, 4, and 6 of life is positively correlated with birth weight, so that larger, healthier infants receive more maternal milk not only earlier but also during each week of hospitalization.

Our results confirm those of previous studies reporting a beneficial effect of maternal milk in reducing sepsis and other infections in VLBW infants. Narayanan et al.21,22 studied low-birth-weight (LBW) (<2.5 kg) infants in New Delhi, India, who were at high risk for infection. The rate of sepsis was significantly reduced in the breastfed group, which included 3 infants with birth weights less than 1500 g. In an intensive care nursery in the United States, El-Mohandes and colleagues12 showed a decrease in the rate of sepsis among LBW infants who received more than 40% of their dietary calories as maternal milk, compared with infants fed preterm formula only. Among those fed any amount of maternal milk, Hylander et al.11 reported a decrease in the number of infections among VLBW infants and in the number and rate of sepsis and meningitis, after controlling for gestational age, 5-minute Apgar score, days of ventilator dependence, and days without oral feeds. No added protective effect with increasing maternal milk intake was noted. Schanler and coworkers30 reported a reduced rate of sepsis and necrotizing enterocolitis in VLBW infants fed at least 30 mL/kg of maternal milk daily compared with preterm formula. These authors also reported an inverse relationship between total volume of maternal milk received and the number of positive blood cultures, which suggests that there is a dose-response effect of maternal milk in reducing sepsis.

Table 3. Protective Effect of Varying Doses of Maternal Milk Through Week 4 of Life, vs No Maternal Milk, on Neonatal Morbidity*

<table>
<thead>
<tr>
<th>Morbidity</th>
<th>0 (n = 40)</th>
<th>1-24 (n = 29)</th>
<th>25-49 (n = 18)</th>
<th>≥50 (n = 32)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sepsis†</td>
<td>10 (25)</td>
<td>13 (45)</td>
<td>9 (50)</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Mean (range)</td>
<td>0.4 (0-3)</td>
<td>0.55 (0-3)</td>
<td>0.56 (0-2)</td>
<td>0.09 (0-2)</td>
</tr>
<tr>
<td>RR (95% CI)‡</td>
<td>1.04 (0.50 to 2.17)</td>
<td>1.17 (0.51 to 2.69)</td>
<td>0.27 (0.08 to 0.95)</td>
<td>0</td>
</tr>
<tr>
<td>NEC (95% CI)§</td>
<td>3 (8)</td>
<td>2 (7)</td>
<td>2 (11)</td>
<td>0</td>
</tr>
<tr>
<td>Retinopathy of prematurity¶</td>
<td>15 (41)</td>
<td>20 (69)</td>
<td>10 (56)</td>
<td>14 (44)</td>
</tr>
<tr>
<td>Chronic lung disease</td>
<td>8 (20)</td>
<td>16 (55)</td>
<td>10 (56)</td>
<td>6 (19)</td>
</tr>
<tr>
<td>OR (95% CI)§</td>
<td>1.78 (0.56 to 5.65)</td>
<td>1.00 (0.27 to 3.75)</td>
<td>1.29 (0.43 to 3.86)</td>
<td>6 (19)</td>
</tr>
<tr>
<td>Jaundice¶</td>
<td>7 (18)</td>
<td>4 (14)</td>
<td>3 (17)</td>
<td>5 (16)</td>
</tr>
<tr>
<td>OR (95% CI)§</td>
<td>3.23 (0.91 to 11.51)</td>
<td>4.95 (1.12 to 22.17)</td>
<td>1.51 (0.37 to 6.24)</td>
<td>1.51 (0.37 to 6.24)</td>
</tr>
<tr>
<td>Ventilator dependence, median (range), d</td>
<td>4 (0-108)</td>
<td>24 (0-50)</td>
<td>18 (0-75)</td>
<td>2 (0.44)</td>
</tr>
<tr>
<td>Difference, log days (95% CI)¶</td>
<td>0.26 (−0.19 to 0.72)</td>
<td>0.12 (−0.40 to 0.64)</td>
<td>−0.02 (−0.46 to 0.41)</td>
<td>0.06 (0.07 to 0.20)</td>
</tr>
</tbody>
</table>

Abbreviations: CI, confidence interval; RR, odds ratio.

*Data are given as number (percentage) unless otherwise indicated.
†Infants with ≥1 episode; analysis compares infants with none vs ≥1 episode.
‡Poisson regression (see “Statistical Analysis” subsection of the “Methods” section). There is a significant difference (at the .05 level) between the group receiving ≥50 mL/kg per day and the reference group (0 mL/kg per day); other differences are not significant.
§Logistic regression (see “Statistical Analysis” subsection of the “Methods” section). Differences between each group and the reference group are not significant at the .05 level.
¶Indirect bilirubin maximum >10 mg/dL (171 µmol/L).
#Mean difference in log days and 95% CI for linear regression (see “Statistical Analysis” subsection of the “Methods” section). Differences between each group and the reference group are not significant at the .05 level.

In this study, we examined the dose effect of maternal milk on neonatal morbidity among VLBW infants. Infants who had received at least 50 mL/kg per day of maternal milk had a significantly lower rate of sepsis than infants who had received no maternal milk. There was no reduction in sepsis among infants receiving lesser amounts of maternal milk. We found no effect of maternal milk on length of hospital stay or other neonatal morbidities, including necrotizing enterocolitis, retinopathy of prematurity, jaundice, chronic lung disease, and duration of ventilator dependence. The mean daily volume of maternal milk received through weeks 2, 4, and 6 of life is positively correlated with birth weight, so that larger, healthier infants receive more maternal milk not only earlier but also during each week of hospitalization.

Table 4. Rates of Sepsis After Weeks 2, 4, and 6 of Life According to Cumulative Daily Volume of Maternal Milk Received Before These Ages

<table>
<thead>
<tr>
<th>Mean Cumulative Daily Volume of Maternal Milk Received, mL/kg</th>
<th>Rate of Sepsis, No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>After Week 2</td>
<td>After Week 4</td>
</tr>
<tr>
<td>(n = 119)</td>
<td>(n = 118)</td>
</tr>
<tr>
<td>0</td>
<td>12/48 (25)</td>
</tr>
<tr>
<td>1-24</td>
<td>15/38 (39)</td>
</tr>
<tr>
<td>25-49</td>
<td>2/20 (10)</td>
</tr>
<tr>
<td>≥50</td>
<td>0/13</td>
</tr>
</tbody>
</table>

*Study infants remaining in hospital: 1 infant was discharged before week 4 of life, and an additional 9 were discharged before week 6.
Maternal milk feeding has been reported to reduce the rate and severity of necrotizing enterocolitis and retinopathy of prematurity in VLBW infants. Because of the low incidence of necrotizing enterocolitis and severe retinopathy of prematurity in this study, a larger sample size would be needed to detect such an effect. If the rate of necrotizing enterocolitis or retinopathy of prematurity was 10%, a sample size of at least 343 infants in each trial arm would be required to detect a 50% decrease in either condition with any certainty (80% power, \( \alpha = 0.05 \), 1-tailed). A significant decrease in length of hospital stay for infants fed maternal milk compared with preterm formula has also been reported; however, our study did not confirm this result.

The mechanism of the anti-infective effect of maternal milk is thought to be via its high immunoglobulin A content and the presence of other immunoactive and immunomodulatory proteins, such as lysozyme, lactoferrin, and cytokines, including interleukins. It is also possible that nonpathogenic maternal bacteria, transmitted via breast milk, colonize the preterm infant gut, thus inhibiting colonization by more pathogenic bacteria or yeast organisms. It has been postulated that skin-to-skin contact, such as kangaroo care, may promote an enteromammary immune response, in which the mother’s exposure to the infant’s nosocomial flora stimulates production of antibodies that are then transmitted to the infant via her breast milk. Neither we nor others have measured the duration of skin-to-skin contact to quantify its potential effect in reducing infection.

Examination of the effect of breast milk on the rate of sepsis among VLBW infants is affected by the method used to quantify maternal milk intake. Few VLBW infants receive all of their oral intake as maternal milk. Breastfeeding has thus been defined for this population in several ways, including receiving a daily mean of more than 50 mL/kg of maternal milk, receiving any human milk feeding, or receiving more than 40% of oral intake as maternal milk. When feeding any maternal milk is considered breastfeeding, infants with disparate intakes are included, possibly obscuring any beneficial effect of maternal milk. When maternal milk intake is measured as a proportion of oral intake, infants receiving primarily intravenous nutrition, but getting all of their oral intake, albeit minimal, as maternal milk, will fall into higher intake categories than infants receiving most oral feeds as maternal milk. For this reason, we measured maternal milk as a proportion of oral and intravenous intake and as a mean volume (in milliliters per kilogram of body weight per day) of maternal milk received. We included daily volumes of 1-24, 25-49, and at least 50 mL/kg and compared these outcomes with those of infants receiving no maternal milk.

The standard of care currently dictates fortification of maternal milk for VLBW infants when the full volume of oral feeds is achieved. As we noted in the “Data Collection” subsection of the “Methods” section, liquid fortifiers double the apparent volume of maternal milk received and, if not excluded for purposes of the analysis, may contribute to an underestimation of the effect of maternal milk on outcomes. Smaller, less mature infants have higher rates of infection. Measurement of the effect of maternal milk on the rate of sepsis should thus be adjusted for birth weight or for time to full oral feeds, to avoid bias in favor of a beneficial effect. Very low-birth-weight infants receive intravenous nutrition for varying lengths of time. Because maternal milk can only be provided orally, smaller and sicker infants who receive less oral intake also receive less maternal milk. This is difficult to completely control for statistically and affects analysis of the effect of maternal milk on neonatal outcomes.

The strengths of this study include its prospective design and the completeness of the nutritional and neonatal data obtained. Weaknesses of the study include the fact that few infants received all of their oral intake as maternal milk and only 27% received at least 50 mL/kg per day, limiting the number of infants in higher categories of maternal milk intake for purposes of analysis. Although we used birth weight and time to full oral feeds as a proxy for illness severity, we did not obtain a risk score such as the clinical risk index for babies or score for neonatal acute physiology, which would have been more useful in this regard. Thirty-two (27%) of our infants received dexamethasone, which has been shown to be associated with decreased rates of infection and may therefore have partially masked the protective effect of maternal milk. Finally, our sample size was not large enough to adequately assess the effect of maternal milk feeding on neonatal morbidities that occur at lower rates, such as retinopathy of prematurity and necrotizing enterocolitis.

In conclusion, the mean number of episodes of sepsis for VLBW infants receiving at least 50 mL/kg per day of maternal milk through week 4 of life was lower by a factor of 0.27 compared with infants receiving no maternal milk. Therefore, a threshold amount of maternal milk was needed to achieve a decrease in the rate of sepsis among VLBW infants, and lower volumes of maternal milk had no demonstrable effect. We could not show an effect of maternal milk on other neonatal morbidities or on length of hospital stay.

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