Influence of changes in lactase activity and small-intestinal mucosal growth on lactose digestion and absorption in preterm infants

Robert J Shulman, William W Wong, and E O’Brian Smith

ABSTRACT

Background: Feeding intolerance (ie, achieving and maintaining full enteral feedings) is a significant problem in preterm infants. A relation exists between feeding intolerance and incomplete lactose digestion.

Objectives: We sought to identify the factors relating to lactose digestion and absorption, lactase activity, and small-intestinal mucosal growth.

Design: Lactose digestion and absorption, lactase-specific activity, and lumen-to-mucosa water flux as a measure of small-intestinal mucosal surface area were determined by using the triple-lumen perfusion technique on 2 occasions 3 wk apart in 10 preterm infants (± SEM gestational age: 28.0 ± 0.2 wk).

Results: Lactose digestion and absorption and lactase activity doubled between studies (P = 0.035 and P = 0.041, respectively). The change in digestion and absorption was related to lactase activity (P = 0.034, R² = 0.38). Lactase activity correlated with gestational age at birth (P = 0.012, R² = 0.51). The number of days of feeding explained 80% of the variability in small-intestinal mucosal surface area (P = 0.001).

Conclusions: To our knowledge, this is the first study to measure directly lactose digestion and absorption, lactase activity, and small-intestinal surface area in preterm infants. Changes in lactose absorption relate primarily to lactase activity rather than to mucosal growth. We showed directly a relation between enteral feeding and small-intestinal mucosal growth.

KEY WORDS Lactose, preterm infant, carbohydrate, small intestine

INTRODUCTION

During development, the greatest increase in lactase activity occurs during the third trimester, when activity rises 3- to 4-fold (1–4). Consequently, preterm infants are lactase deficient (1). As would be anticipated from these data, most studies have shown that the digestibility of lactose is incomplete in preterm infants (1, 5, 6).

Feeding intolerance (ie, achieving and maintaining full enteral feedings) is a serious problem in preterm infants (7). Indeed, a relation exists between the time required to reach full enteral feedings and the duration of hospitalization (8). Recent studies have shown a relation between feeding intolerance and dietary lactose–lactase activity in preterm infants. A greater intake of lactose appears to increase the risk of feeding intolerance, and the likelihood of feeding intolerance is inversely related to lactase activity (3, 9). Similarly, increasing the intake of lactose may adversely affect weight gain (10).

Other studies and observations have drawn a relation between lactose malabsorption and the development of intestinal disease in preterm infants. Studies by Kien (11) defined the potential role of lactose malabsorption in the development of necrotizing enterocolitis. Another recent study in preterm infants showed that lactose malabsorption in some infants causes an increase in fermentative products (ie, short-chain fatty acids) and a shift in the profile of short-chain fatty acids (eg, increased butyric acid) arising from colonic bacterial metabolism of the lactose (12). The increased butyric acid production was associated with bloody stools and abdominal distention (12). Butyric acid in high concentrations has been shown to be toxic to colonic mucosa in cell lines and animal models (13, 14). This fits with the recent observation in an animal model of necrotizing enterocolitis that the appearance of intestinal lesions requires a combination of low endogenous lactase activity, lactose in the diet, and colonization by lactose-fermenting bacteria (15).

Despite the apparent importance of dietary lactose and lactase activity in feeding preterm infants, gaps remain in our knowledge of the factors that influence the development of lactose digestion and absorption and lactase activity in preterm infants. Previous work in miniature pigs suggested that age-related changes in lactose digestibility are related not only to lactase-specific activity but also to changes in small-intestinal mucosal growth (ie, surface area) (16). We hypothesized that the same would be true for preterm infants. Our objective was to determine the contribution of each of these (lactase-specific activity and mucosal growth) to the maturational change in lactose digestion and absorption. Our second objective was to identify demographic and

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clinical variables that might influence the expression of lactase-specific activity and small-intestinal mucosal growth.

To investigate our hypothesis, we took advantage of the ability to measure lactase-specific activity in intestinal epithelial cells collected during intestinal perfusion studies. The perfusion studies allow us to measure lactose digestion and absorption concurrently as well as small-intestinal mucosal surface area by using the lumen-to-mucosa flux of deuterium-labeled water in a prospective manner.

SUBJECTS AND METHODS

Design

We measured directly lactose digestion and absorption, net water absorption, lactase-specific activity, and unidirectional lumen-to-mucosa water flux in preterm infants by using the triple-lumen perfusion catheter technique (described below). Infants were studied when they were tolerating orogastric feedings with the triple-lumen perfusion catheter technique (described below). Infants were studied when they were tolerating orogastric feedings and no blood in stools), and the attending physician considered them to be stable. The measurements were repeated 3 wk later to assess changes in lactose digestion and absorption, water absorption, lactase activity, and unidirectional lumen-to-mucosa water flux. Infants were selected for study if they were born at ≤30 wk gestation, were appropriate for gestational age, and had no history of gastrointestinal, renal, cerebral, or chronic pulmonary disease (ie, the neonatal course was uneventful). On the basis of our previous studies, we anticipated that we would have to study ≥10 infants to detect significant changes over time (5, 17, 18). Informed consent was obtained from the parents. The study was approved by the Baylor College of Medicine Institutional Review Board for Human Research.

We chose the time period of 3 wk for several reasons. The few available studies of postmortem specimens taken from preterm infants suggest that lactase activity increases significantly over this period (19–21). Although to our knowledge no data are available regarding the change in small-intestinal mucosal surface area during this gestational age period, there is a measurable increase in small-intestinal length as determined from postmortem studies (22–25). The final consideration was a practical one. A longer time period between perfusions would have been ideal. However, by waiting it was more likely that the infant would not be able to undergo the second study. Placement of the perfusion catheter is only done in infants who are already receiving enteral feedings. It is not appropriate to insert a catheter in an otherwise healthy infant who no longer has a feeding tube.

Two perfusions were carried out on each study day: one to measure lactose digestion and absorption and lactase activity and the other to determine net water absorption and unidirectional lumen-to-mucosa water flux (ie, small-intestinal mucosal surface area). On each study day, the infants underwent perfusions 1 h after their previous feeding. After any residual feeding was suctioned out, the orogastric feeding tube was removed and the perfusion catheter was placed (see below). Infants received 2 solutions administered in random order: one contained lactose and the other deuterated water ($D_2O$) (Table 1).

Perfusion study

To place the triple-lumen perfusion catheter safely in the premature infants without the use of X-rays, we constructed the catheter from two 6 French feeding tubes with a pH-sensitive electrode at the tip, thus allowing immediate pH readings (Acusite pH Enteral Feeding System; Zinetics Medical, Salt Lake City). One feeding tube provided the infusion port and the other tube was used for the distal collecting port. A 5 French feeding tube (Argyle Inc, St Louis) attached to the two 6 French tubes provided the proximal collecting port. The infusion port and the 2 collecting ports (proximal and distal) were spaced 10 cm apart. By monitoring the change in pH, we could verify the location of the perfusion catheter. The tube was considered to be positioned properly when the perfusing port pH changed from <4.0 (gastric) to >5.5 (duodenal). On the basis of our previous experience, the perfusing port is located in the third to fourth portion of the duodenum and the distal collecting port in the proximal jejunum (5, 17, 18).

Lactose absorption was determined as described previously (5, 17, 26). In brief, the method is based on the changes in concentration of a test substance (lactose) that occur in the lumen of the intestine relative to polyethylene glycol (PEG) 4000. Because PEG 4000 is not absorbed significantly, the amount infused per unit time is equal to the amount reaching the proximal and distal collecting ports. This allows calculation of water and solute transport (26):

\[
V_2 = (V_1 \times M_1/M_2) - E_p \quad (1)
\]

\[
V_3 = V_2 \times M_2/M_1 \quad (2)
\]

\[
\Delta H_2O = V_2 - V_3 \quad (3)
\]

\[
\Delta S = V_2 \times S_2 - V_3 \times S_3 \quad (4)
\]

where $V_1$ is the infusion rate; $V_2$ is the flow rate between the infusion and proximal collecting ports; $V_3$ is the flow rate between the proximal and distal collecting ports; $E_p$ is the rate of aspiration at the proximal port; $M_1$, $M_2$, and $M_3$ are the PEG 4000 concentrations in the perfusion solution, at the proximal port, and at the distal port, respectively; $\Delta H_2O$ is the rate of water transport between the middle and distal collecting ports; $S_2$ and $S_3$ are the lactose concentrations at the proximal and distal collecting ports, respectively; and $\Delta S$ is the rate of lactose digestion and absorption between the middle and distal collecting ports.

The perfusate solution was maintained at 37 °C. The rate of perfusion was 1.0 mL/min. A 45-min equilibration period was used, followed by 30-min collections from each port. Collections from the distal collection port began 5 min after collections from the proximal collection port to allow for transit between the ports (5, 17, 18). Samples were collected on ice and were then immediately frozen in liquid nitrogen and stored at −70 °C until analyzed.

<table>
<thead>
<tr>
<th>TABLE 1 Composition of perfusion solutions</th>
<th>Lactose solution</th>
<th>$D_2O$ solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>NaCl (g/L)</td>
<td>2.2</td>
<td>7.9</td>
</tr>
<tr>
<td>PEG 4000 (g/L)</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Lactose (g/L)</td>
<td>70</td>
<td>—</td>
</tr>
<tr>
<td>$D_2O$ (ppm)</td>
<td>—</td>
<td>35 000</td>
</tr>
<tr>
<td>Osmolality (mOsmol/L)</td>
<td>270</td>
<td>270</td>
</tr>
</tbody>
</table>

Note: $D_2O$, polyethylene glycol.
The collected effusate was analyzed as described previously for volume, for PEG 4000 concentration (by the cold acetone precipitation method), and for lactose by using a commercial kit (Boehringer Mannheim GmbH, Mannheim, Germany) (5, 17). The deuterium enrichment of the effusate was analyzed by gas isotope ratio mass spectrometry. Ten microliters of effusate without further treatment was reduced to hydrogen gas with 200 mg Zn reagent at 500 °C for 30 min (27). The δ²H:¹H isotope ratios of the hydrogen gas were measured with a Finngan Delta-E gas isotope ratio mass spectrometer (Finngan MAT, San Jose, CA). The results are expressed in delta (δ) per mil (‰) units, which are defined as follows:

\[ \delta^2H (\text{‰}) = \left( \frac{R_s/R_i - 1}{X} \right) \times 10^3 \]  

where \( R_s \) and \( R_i \) are the \( ^2H:¹H \) isotope ratios of the sample and standard, respectively. The \( \delta^2H \) values were normalized against 2 international water standards: Vienna-standard mean ocean water and standard light Antarctic precipitation (28).

Lactase activity was measured in brush border membranes prepared from the effusate. Torres-Pinedo et al (29, 30) observed that luminal fluid obtained from intestinal perfusion studies in infants contains sediment consisting of whole epithelial cells and brush borders. They showed that it is possible to measure protein and disaccharidase activity in the recovered material (29, 30). Similar techniques have been used in patients with inflammatory bowel disease to characterize intestinal epithelial cells (31). Studies by Aramayo et al (32) also showed that there is a strong correlation in infants and children between disaccharidase activity measured in small-intestinal mucosal biopsies and in the adjacent intestinal luminal fluid. Luminal lactase activity was found to increase with age (33). In a more recent study, the investigators showed even more convincingly that measured luminal lactase activity reflects that in the adjacent intestinal mucosa (34). Consequently, we used this method to measure lactase activity in the effusate. In brief, the effusate was centrifuged at 4 °C at 1600 × g for 10 min. The pellet was reconstituted to the original volume with tris:HCl:mannitol buffer. CaCl₂ (100 mmol/L) was added at a concentration of 10% of the ending volume, and the samples were incubated at 4 °C with mixing for 15 min. The samples were then centrifuged at 4 °C at 1600 × g for 10 min. The supernatant fluid was spun at 3000 × g at 4 °C for 30 min. The pellet was reconstituted with 1 mL of tris:HCl:mannitol buffer. The microvillus membranes were frozen at −70 °C until analyzed for lactase activity and protein. Lactase activity and protein concentration (Pierce Chemical Co, Rockford, IL) in the brush border membranes were analyzed as described previously (35, 36).

Calculations

Net lactose digestion and absorption were calculated as previously described (5, 17, 26). Lactose digestion and absorption were calculated as the difference between the total amount of lactose, glucose, and galactose at the beginning and end of the test segment (ie, between the middle and distal collecting ports). Lactase activity was expressed as specific activity (μmol · min⁻¹ · g protein⁻¹).

Water absorption was calculated as previously described (5, 17, 18, 26). Unidirectional water (ie, \( D_2O \)) absorption as a measure of intestinal surface area was calculated by using the method of Fordtran et al, Lewis and Fordtran, and Fiedorek et al (37–39). Fordtran et al (37, 38) showed in adults that labeled water could be used to measure unidirectional water absorption with the intestinal perfusion technique. In brief, the net volume change in the lumen of the perfused segment of intestine is calculated by using the changes in the nonabsorbable marker PEG 4000. The net volume change is a result of fluxes of water in and out of the lumen. The fluxes are measured by adding labeled water (\( D_2O \)) to the perfusate (37, 38).

\[ ^2H \times \text{volume infused} = \text{total } ^2H \text{ infused} \]  

\[ ^2H \text{ remaining} \times \text{volume remaining} = \text{total } ^2H \text{ remaining} \]  

\[ \text{Total } ^2H \text{ infused} - \text{total } ^2H \text{ remaining} = \text{total } ^2H \text{ absorbed} \]  

\[ \text{Total } ^2H \text{ absorbed} = \text{total } ^2H \text{ infused} \times \text{volume infused} \]  

\[ \text{flux of water out of lumen} \]

Lewis and Fordtran (38) showed in a rat model that unidirectional water flux out of the lumen correlates well with maximal glucose transport rates, which, in other studies, were also shown to reflect intestinal mucosal surface area (40, 41). Fiedorek et al (39) carried out rat experiments in which the unidirectional water flux out of the lumen (using labeled water) was shown to correlate closely with surface area as measured directly by computer-assisted planimetry and mucosal weight \( (R^2 = 0.79) \). For simplicity, we use the term small-intestinal mucosal surface area.

Statistics

Results are given as means ± SEMs. Comparisons of the results of the studies carried out 3 wk apart were done by using a one-tailed paired \( t \) test given that the changes over time should be in one direction (ie, increased). Linear regression analysis was used to test for relations between lactose absorption, lactase-specific activity, small-intestinal mucosal surface area, and either demographic variables or variables related to feeding history (correlates with demographic and clinical characteristics) after testing to ensure that the data were distributed normally. Spearman rank correlation was also used as a more conservative approach. \( P \) values < 0.05 were considered significant. Minitab 14 (Minitab Inc, State College, PA) was used for the statistical analyses.

RESULTS

A total of 19 infants were enrolled into the study. Of these, 10 infants had complete perfusion studies done 3 wk apart (ie, complete studies). The reasons some of the infants did not have 2 complete perfusions studies done included parental request that the infant be withdrawn from the study, lack of permission from the attending physician because of concern that the infant was not stable enough, and inability to obtain adequate volume of samples during the perfusion.

The clinical characteristics of the infants are given in Table 2. Three of the infants received a 24-kcal/oz formula for preterm infants (Enfamil Premature 24; Mead-Johnson, Evansville, IN), and the remaining 7 received their mother’s milk. Mothers of 6 of
the 10 infants received antenatal glucocorticoids. None of the infants received postnatal steroids.

Perfusion results

Lactose digestion and absorption and lactase activity increased significantly between studies (Figure 1). In contrast, although the mean values were numerically greater for the second study, the changes in net water absorption and small-intestinal mucosal surface area were not significant.

There was a significant relation between the change in lactose digestion and absorption over the 3-wk period and lactase activity at the time of the second study (Figure 2). In 2 infants, lactose digestion and absorption appeared to decrease slightly over the 3-wk period. Most likely this was within the variation of the method and actually reflected no change in lactose digestion and absorption over the 3-wk period.

The relation between the change in lactose digestion and absorption and that in lactase activity over the 3-wk period was not significant \( (P = 0.17) \) because of one outlier (data not shown). The change in lactose digestion and absorption did not correlate with small-intestinal mucosal surface area at either the first or the second study.

Correlates with demographic and clinical characteristics

We tested potential demographic and clinical characteristics that might be related to lactase-specific activity, small-intestinal mucosal surface area, or both. These included birth weight, gestational age, the age at which feedings began, the type of

<table>
<thead>
<tr>
<th>Variable</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gestational age (wk)</td>
<td>28 ± 0.2</td>
</tr>
<tr>
<td>Birth weight (g)</td>
<td>1002 ± 62</td>
</tr>
<tr>
<td>Age at which feedings began (d)</td>
<td>9.6 ± 1.3</td>
</tr>
<tr>
<td>Initial feeding amount (mL · kg(^{-1} ) · d(^{-1}))</td>
<td>10.6 ± 2.1</td>
</tr>
<tr>
<td>Age at first study (d)</td>
<td>27 ± 3</td>
</tr>
<tr>
<td>Weight at first study (g)</td>
<td>1368 ± 92</td>
</tr>
<tr>
<td>Days of feeding to first study (d)</td>
<td>14.7 ± 2.0</td>
</tr>
<tr>
<td>Amount of feeding at first study (mL · kg(^{-1} ) · d(^{-1}))</td>
<td>113 ± 12</td>
</tr>
<tr>
<td>Age at second study (d)</td>
<td>50 ± 4</td>
</tr>
<tr>
<td>Weight at second study (g)</td>
<td>1901 ± 116</td>
</tr>
<tr>
<td>Days of feeding to second study (d)</td>
<td>37.2 ± 4.6</td>
</tr>
<tr>
<td>Amount of feeding at second study (mL · kg(^{-1} ) · d(^{-1}))</td>
<td>140 ± 6</td>
</tr>
</tbody>
</table>

\(^{1}\) All values are \( \bar{x} \pm \text{SEM}; n = 10. \
feeding (human milk or formula), the total volume of full-strength feedings the infant had received, the volume of feedings the infants were receiving at the time of the studies, and administration of antenatal glucocorticoids.

Demographic and clinical characteristics versus lactase activity

Lactase-specific activity at the time of the second study correlated with gestational age at birth (Figure 3). There appeared to be an inverse relation between lactase-specific activity at the time of the second study and the age at which feedings were started, but this did not quite reach significance (Figure 3). There was no relation between lactase-specific activity at the time of the second study and the other demographic and clinical characteristics. Lactase-specific activity at the time of the first study did not correlate with any demographic or clinical characteristic.

There was a significant relation between the change in lactase-specific activity between studies and birth weight (Figure 3). One infant had a small decline in lactase activity, the reason for which is not clear. This may have reflected a true variation in activity between studies, slightly different placement of the perfusion catheter between studies, some other unknown variable, or a combination.

The change in lactase-specific activity did not correlate as well with gestational age ($P = 0.057, R^2 = 0.30$; data not shown). The change in lactase activity between studies versus the age at which feedings were started was not significant, although there was a trend ($P = 0.086, R^2 = 0.24$; data not shown).

Demographic and clinical characteristics versus small-intestinal mucosal surface area

There was a strong relation between small-intestinal mucosal surface area at the time of the first study and the number of days the infant had been fed by the time of the first study (Figure 4). There appeared to be a relation between small-intestinal mucosal surface area at the time of the first study and the number of days the infant had been fed full-strength formula (Figure 4). There were no correlations between other demographic or clinical characteristics and small-intestinal mucosal surface area at the time of the first study.

Similar to what was found for the first study, there appeared to be some relation between small-intestinal mucosal surface area at the time of the second study and the number of days the infant had been fed full-strength formula to that point ($P = 0.057, R^2 = 0.34$; data not shown). There were no correlations between other
Previously adapted the triple-lumen intestinal perfusion technique for the clinical course of the infant (1, 2, 3, 19–21). Having previously obtained postmortem specimens (ie, not sequentially) with little information on activity, and small-intestinal mucosal surface area. We also were limited for the first time to relate these gastrointestinal developmental changes to demographic or clinical characteristics. Limited direct data are available regarding the developmental increase in lactase activity in preterm infants. The previous data were obtained from indirect measures or from a limited number of autopsy specimens (ie, not sequentially) with little information on the clinical course of the infant (1, 2, 3, 19–21). Having previously adapted the triple-lumen intestinal perfusion technique for direct data are available regarding the developmental increase in lactose digestion and absorption, lactase activity increases with age out of proportion to the increase in small-intestinal mucosal mass. The increase in lactase activity over the 3-wk study period was greater than the increase in small-intestinal mucosal mass (Figure 1). Our results suggest that changes in lactose digestion and absorption with age likely are related more to an increase in lactase-specific activity than to small-intestinal mucosal growth (Figures 1 and 2).

Factors in lactase development

Lactase is the sole enzyme responsible for the digestion of lactose. Lactase activity could increase via an increase in lactase-specific activity (ie, lactase protein production, decreased breakdown, or both), via an increase in small-intestinal mucosal mass (ie, the amount of lactase protein does not change per unit of total protein, rather the total mass of lactase increases as a consequence of, and in proportion to, small-intestinal mucosal growth), or both. The current data support the idea that lactase-specific activity increases with age out of proportion to the increase in small-intestinal mucosal mass. The increase in lactase activity over the 3-wk study period was greater than the increase in small-intestinal mucosal mass (Figure 1). Our results suggest that changes in lactose digestion and absorption with age likely are related more to an increase in lactase-specific activity than to small-intestinal mucosal growth (Figures 1 and 2).

A previous study in neonatal, young, and adult pigs by Redel et al (16) suggested that in this species, lactase-specific activity, in contrast with mucosal mass, also was the primary determinant of lactose digestion and absorption. Consequently, we hypothesize that attempts to increase lactose digestion and absorption (and thereby reduce the risk of feeding intolerance) are more likely to succeed by strategies that specifically facilitate the development of lactase-specific activity as opposed to increasing nonspecifically small-intestinal mucosal surface area.

Factors in lactose digestion and absorption

Lactase malabsorption was previously shown to impair the health of preterm infants, leading to feeding intolerance, delayed attainment of full enteral feedings, and poor weight gain (3, 9, 10). There is evidence that it may play a role in the development of necrotizing enterocolitis (11–13). Thus, it is important to understand the processes involved in the maturation of lactose absorption in preterm infants. Interestingly, one report suggested that even in some full-term infants lactose malabsorption may cause colic-like symptoms (42).

DISCUSSION

These results are the first to measure simultaneously the age-related increase in lactose digestion and absorption, lactase activity, and small-intestinal mucosal surface area. We also were able for the first time to relate these gastrointestinal developmental changes to demographic and clinical characteristics. Limited direct data are available regarding the developmental increase in lactase activity in preterm infants. The previous data were obtained from indirect measures or from a limited number of autopsy specimens (ie, not sequentially) with little information on the clinical course of the infant (1, 2, 3, 19–21). Having previously adapted the triple-lumen intestinal perfusion technique for use in small preterm infants, we could obtain lactase measurements sequentially in the same infant and relate enzyme activity to other physiologic, demographic, and clinical characteristics (5, 17, 18).

Lactase malabsorption was previously shown to impair the health of preterm infants, leading to feeding intolerance, delayed attainment of full enteral feedings, and poor weight gain (3, 9, 10). There is evidence that it may play a role in the development of necrotizing enterocolitis (11–13). Thus, it is important to understand the processes involved in the maturation of lactose absorption in preterm infants. Interestingly, one report suggested that even in some full-term infants lactose malabsorption may cause colic-like symptoms (42).

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Factors in lactase development

In the current study, we were able to examine some of the possible factors related to the development of lactase activity. Activity was correlated with the maturational state of the infant at birth (ie, gestational age; Figure 3). Given that lactase-specific activity appears to be governed, in part, by the maturational state of the infant at birth, it is tempting to speculate that variations in lactase activity in individual infants at birth may help to explain why feeding tolerance can vary widely in infants of the same gestational age (3).

Indirect measures have suggested a relation between feeding and the development of lactase activity. In preterm infants, we showed by using the oral administration of lactulose and lactose and subsequent measurement of these sugars in the urine that feeding can enhance lactase activity at 28 d of age (3). McClure et al (2) corroborated these findings by using lactase:sucrase ratios measured in duodenal fluid aspirates. Although the relation only showed a trend in our current study (likely because of the smaller number of infants than in the previous study), it appears that the current results support this observation. Lactase-specific activity at the time of the second study as well as the
change in lactase-specific activity between studies bore some relation with the time at which enteral feedings were started.

Factors in small-intestinal mucosal growth

To our knowledge, no studies have measured small-intestinal mucosal growth in vivo in preterm infants. Several reports using necropsy material have described the increase in small-intestinal length that occurs in utero (22–25). It would be anticipated that intestinal mucosal surface area and intestinal length would be correlated, and this appears to be the case (16). Small-intestinal length increases linearly in relation to gestational age (22–25). Indeed, in our study, small-intestinal mucosal surface area as measured by the unidirectional lumen-to-mucosa flux of D2O increased arithmetically over the 3-wk study period, although the increase was not significant (Figure 1). This likely was due, in part, to the relatively short time (3 wk) between studies and possibly to the variation inherent in the perfusion technique.

Feeding appeared to have a significant relation with small-intestinal mucosal surface area. It correlated strongly with the number of days the infants had received feedings before the first study (Figure 4). This explained 80% of the variability in small-intestinal mucosal surface area (Figure 4). Similarly, we observed a potential relation with the total days of full-strength feedings the infant had received at the time of the study (Figure 4). These observations suggest that feeding does enhance small-intestinal mucosal growth in human infants. To our knowledge, this is the first direct observation of the relation between feeding and small-intestinal mucosal growth in human infants.

Certain caveats should be kept in mind regarding the interpretation of our results. Unfortunately, we could not study a larger number of infants. Thus, some of the changes over time might have been greater or the relations between variables may have been more robust with a larger group of infants. Even though the placement of the perfusion catheter allows measurement of lactose absorption and lactase activity in the proximal jejunum, where lactase activity is highest (1, 19–21), this may not reflect completely the totality of changes occurring throughout the small intestine. Although previous work on measuring small-intestinal surface area with the use of unidirectional water flux validates the use of this method, it is possible that the method provides only an estimate of surface area rather than a perfect measure (37–39).

Changes in blood flow to the gastrointestinal mucosa may affect the estimate of surface area rather than a perfect measure (37–39). Changes in blood flow to the gastrointestinal mucosa may affect unidirectional water flux (43). Given that the infants were studied in the same manner, this is unlikely to have played a major role. However, this may explain, in part, some of the variability in the measurements. Finally, although lactose malabsorption may play a role in feeding intolerance, it remains to be determined what steps can be taken to safely reduce any potential risk.

Summary

In preterm infants, maturational changes in lactose digestion and absorption appear to be related more to changes in lactase-specific activity than to changes in small-intestinal mucosal surface area. Lactase-specific activity increases out of proportion to that which can be explained solely by changes in small-intestinal mucosal growth. Lactase-specific activity is related to the maturational state of the infant and to some degree to enteral feeding. In contrast, small-intestinal mucosal surface area is related primarily to enteral feeding. We thank S Henning for helpful comments. RJS was responsible for the design of the study, placing the perfusion catheters, and overseeing the perfusions and analyses. Michelle Malanga carried out the bedside collections and obtained the clinical and demographic information. WWW carried out the water flux analyses. Mary Ann Rauch performed the other analyses. EOS assisted in statistical analyses. None of the authors had any financial or personal interest in any company or organization sponsoring the research.

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