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Responses of Gastrointestinal Peptides and Motor Activity to Milk and Water Feedings in Preterm and Term Infants

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ABSTRACT. Because duodenal motor activity differs between preterm and term infants during fasting, this study evaluated the responses of motor activity and peptide release in response to feeding. In the first study, fasting concentrations of gastrin, gastric inhibitory peptide, neurotensin, and peptide YY (PYY) were determined in 53 preterm and 20 term infants. Plasma concentrations of gastrin and neurotensin were significantly lower in preterm infants than in healthy adults reported previously by our lab ($p < 0.01$). Plasma concentration of gastric inhibitory peptide and PYY were higher than in healthy adults ($p < 0.01$). Gastrin concentrations in preterm and term infants varied directly with gestational age ($p < 0.005$); PYY varied inversely with gestational age ($p < 0.005$). In a secondary study, intestinal manometry was recorded and serial peptide concentrations were determined in 43 preterm babies who were given their first enteral feeding intraduodenally with formula or sterile water. Although none of the four peptide plasma concentrations changed in response to feeding with water, plasma concentrations of gastric inhibitory peptide, neurotensin, and PYY significantly increased with formula feedings ($p < 0.05$ or less). In addition, plasma gastrin increased significantly in seven infants fed milk compared with eight fed water by orogastric tube ($p < 0.01$). In contrast to the peptide response to feeding, motor activity changed in response to feeding with either water or milk; motility indices increased and periods of motor quiescence decreased significantly during feeding as compared with fasting ($p < 0.02$). Responses of both motor activity and peptides to feeding were time related. Although fasting concentrations of four regulatory peptides were immature in preterm infants compared with adults, postprandial responses to nutrient feedings were present in the first days of life. This discrepancy in functional maturation of the preterm intestine during fasting and feeding is present for both motor activity and peptide response, and we speculate that the controlling mechanisms of these two phases of digestion may mature independently at different postconceptual ages. (*Pediatr Res* 31: 587–590, 1992)

Abbreviations

GIP, gastric inhibitory peptide
NT, neurotensin
PYY, peptide YY
NICU, Newborn Intensive Care Unit

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Motor activity, which is responsible for the aboral movement of nutrients through the gastrointestinal tract is modulated by neural and hormonal influences. We have recently shown that fasting intestinal motor activity differs between preterm and term infants (1) and, in turn, between infants and adults. Despite the presence of these differences in motor activity during fasting, the motor responses to feeding are similar in preterm and term infants (2). In a parallel fashion, fasting intestinal hormone concentrations differ between preterm infants and adults (3). However, hormonal responses to feeding in preterm and term infants are present, although blunted or overly reactive, in the 1st postnatal wk when cross-sectional sampling techniques are used (3, 4). No previous study in preterm infants has validated that hormonal responses to feeding are present in neonates by using serial sampling of plasma concentrations of peptides. Inasmuch as motor responses to enteral feedings are present in preterm infants, we hypothesized that peptide responses to feedings would also be present. The purpose of the preliminary study was to determine plasma concentration of four gastrointestinal hormones and peptides in preterm and term infants and to assess the influence of gestational maturation on their concentration. The purpose of the secondary study was to compare serial peptide concentrations and motor activity in infants fed water or artificial formula to determine if peptide concentrations change in response to enteral feeding.

SUBJECTS AND METHODS

First study: levels of peptides during fasting. Study participants in this first study were 63 preterm and 20 term infants admitted to Saint Mary's Hospital NICU. All infants were admitted for oxygen or ventilator support for respiratory disease, including respiratory distress syndrome, congenital pneumonia, or meconium aspiration. All infants had indwelling central venous or arterial catheters for routine NICU support and monitoring. Preterm infants ranged from 25 to 35 wk of gestational age, and term infants from 36 to 42 wk (Table 1). Preterm infants participated in this study on postnatal d 3.8 and term infants on d 4.1 (Table 1). By NICU routine, all babies were receiving i.v. fluids and had not received any enteral feedings. This protocol was approved by the Mayo Clinic Institutional Review Board, and parents provided informed consent for their infants' participation in this study. A single 2-mL blood sample was withdrawn from each infant's indwelling central catheter. Blood was placed in tubes containing 1.5 mg of EDTA per mL of whole blood.

Table 1. *Patient characteristics and comparison of fasting plasma gastrointestinal peptide concentrations in preterm and term infants and adults (mean ± SEM)*

	Preterm	Term	Adults*
n	63	21	15
Gestational age (wk)	30.9 ± 0.3	37.7 ± 0.4	
Postnatal age (d)	4.2 ± 0.2	5.4 ± 0.7	
Birth weight (g)	1570 ± 74	3167 ± 189	
Peptide concentrations (ng/L)			
Gastrin	59 ± 4†	82 ± 9†‡	100 ± 10†‡
GIP	578 ± 36†§	588 ± 70†§	230 ± 26†
NT	27 ± 4†§	32 ± 6†§	67 ± 8†
PYY	748 ± 64†§	530 ± 64†§	75 ± 9†

* Previously reported in Ref. 5.
† $p < 0.0001$, analysis of variance among age groupings.
‡ Differ from preterms, Newman-Keuls test.
§ Differ from adults, Newman-Keuls test.

Samples were centrifuged, and the plasma was stored at −20°C. RIA was used to determine in each sample the concentration of gastrin, GIP, NT, and PYY. Duplicate 100-μL samples were used for each assay for gastrin, NT, and PYY; 200-μL samples were used for GIP. The characteristics of these assays have been previously published (5). In brief, all assays had an intraassay variation of 8% or less and all plasma concentrations of infants fell within the range of absolute sensitivity for each assay used.

Feeding study. In this secondary study, 58 additional preterm infants participated as subjects; 43 received duodenal feedings and another 15 received gastric feedings. Infants ranged from 27 to 33 wk of gestational age and had been admitted for respiratory distress syndrome. All were receiving parenteral nutrition and had received no enteral feedings. All were studied by postnatal d 3. This protocol was also approved by the Mayo Clinic Institutional Review Board, and parents provided informed consent for their infant's participation in this study. Each infant was studied on the day that enteral feedings were to be initiated. Motor activity was evaluated using a neonatal manometric system that has been previously described and validated (6). In short, this system is a low-compliance, continuous perfusion system that provides a response rate of 57 mm Hg/s at 10 psi at a flow rate of 0.01/mL/min/recording port (6). Before any feedings were given, a 3.5-mm motility/feeding tube was placed in the upper duodenum and the position verified by the presence of motor activity characteristic of the duodenum. Motor activity was recorded for 4 h. At the end of 4 h, blood was collected for determination of regulatory peptide concentrations. Each of 43 infants was then randomly assigned to receive a feeding with sterile water or Similac formula intraduodenally via the motility/feeding tube. All infants were fed 4 mL/kg as a 2-h continuous infusion. Blood samples of 1.2 mL were withdrawn from each infant's indwelling central catheter at 30, 60, 150, and 180 min. Blood was processed and peptide concentrations determined as for the preliminary study. The additional 15 infants were fed 4 mL/kg feeding of milk ($n = 7$) or water ($n = 8$) by orogastric tube, and serial sampling was performed as described for infants fed by oroduodenal tube.

Data analysis. In the first study, plasma concentrations of all four peptides were compared among preterm infants, term infants, and adults by analysis of varience, with Newman-Keuls testing to identify the presence of intergroup differences. Regression analysis was performed for each peptide to assess peptide concentration in infants as a function of gestational age. Significant differences were identified when p was less than 0.05. In the secondary study, plasma concentrations of peptides at 30–180 min were determined, and the peptide release during feeding was quantified by determining an integrated area under the curve for each peptide using the trapazoid rule. These integrated areas were compared between water- and milk-fed infants by unpaired t test. Data analysis for babies fed duodenally and intragastrically was performed separately and expressed as mean ± SEM.

In the manometric study, motor activity was analyzed in 30-min segments. Quiescence was defined as an absence of motor activity. Duration of quiescence was expressed as min of quiescence/h of recording/recording lead. Motility index, a quantification to identify the presence of the fed response, was calculated as $\log_e$ (sum of amplitude × number of pressure waves + 1) (7). Quiescence duration and motility index were compared during fasting and feeding infusion by paired t test. To further characterize the feeding response (2), the number of pressure waves/30 min were calculated for each 30-min interval throughout the 180-min study and each value was compared with that for time 0 by paired t test.

RESULTS

Preliminary study of peptide levels during fasting. Fasting plasma concentrations of all four hormones and regulatory peptides differed among preterm infants, term infants, and healthy nonpregnant females previously reported by our laboratory (5), (Table 1; all $p < 0.0001$). Gastrin concentrations were lower in preterm infants than in term infants and adults, whereas NT concentrations were lower in both preterm infants and term infants compared with adults. GIP and PYY concentrations were significantly higher in preterm and term infants than in adults. Gastrin concentrations varied directly with gestational age ($r = 0.33$; $p < 0.005$). GIP and NT did not vary with gestational age. PYY varied inversely with gestational age ($r = -0.38$; $p < 0.001$).

Peptide responses to feeding. There was no significant difference in plasma concentrations in gastrointestinal peptides during fasting in water and milk-fed infants (Table 2). Water- and milk-fed infants differed with respect to their hormonal response to feeding. Plasma gastrin did not significantly change throughout the 3-h study period in either milk- or water-fed infants, and their integrated responses were similar (Fig. 1). In contrast to gastrin, the integrated response of GIP, NT, and PYY were significantly greater in milk-fed infants compared with water-fed infants (all $p < 0.05$ or less, Fig. 1).

In contrast to the infants fed by intraduodenal infusion, gastrin

Table 2. *Characteristics of preterm infants fed water or milk (mean ± SEM)*

	Water	Milk	*p*
n	21	22	
Gestational age (wk)	30.6 ± 0.5	31.5 ± 0.5	NS
Birth weight (g)	1552 ± 117	1725 ± 126	NS
Postnatal age (d)	4.9 ± 0.5	5.3 ± 0.5	NS
Fasting plasma concentration (ng/L)			
Gastrin	59 ± 6	60 ± 7	NS
GIP	533 ± 54	641 ± 59	NS
NT	19 ± 3	31 ± 7	NS
PYY	793 ± 100	566 ± 84	NS

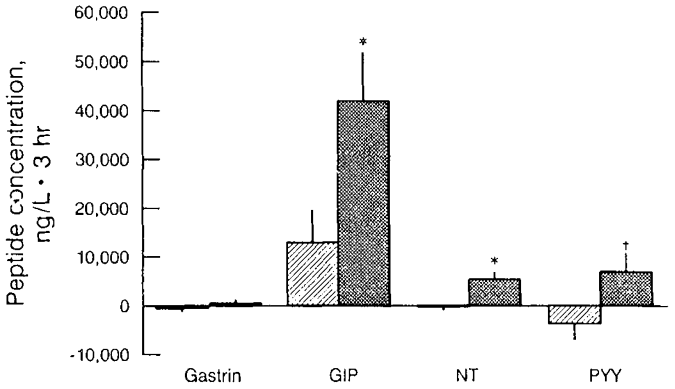

Fig. 1. Integrated concentrations of gastrointestinal peptides in preterm infants fed water or formula by oroduodenal tube (mean ± SEM). Infants fed water are represented by *bars with diagonal cross-hatching* and infants fed formula, by *stippled bars*. Significant differences are designated by * ($p < 0.05$) and † ($p < 0.01$).

Table 3. *Motor activity during fasting and feeding in water and milk-fed preterm infants (mean ± SEM)*

	Fasting	Fed	*p*
Water-fed			
Motility index*	12.6 ± 0.2	13.3 ± 0.2	0.001
Quiescence duration (min/h/lead)	22.7 ± 1.8	18.0 ± 2.6	0.02
Milk-fed			
Motility index*	12.6 ± 0.2	13.3 ± 0.1	0.001
Quiescence duration (min/h/lead)	23.5 ± 1.8	17.7 ± 1.9	0.02

* Motility index, sum of $\log_e$ (sum of amplitudes × number of pressure waves + 1).

concentrations differed with milk or water feedings when infants were fed by orogastric tube. Plasma gastrin did not differ over 180 min in eight water-fed infants. In contrast to water-fed infants, plasma gastrin in seven milk-fed infants was significantly higher at 30 and 60 min compared with fasting, and the integrated gastrin response to milk feeding was significantly greater than that to water feeding (137 ± 100 *versus* 1260 ± 320 pg/mL·1 h; $p < 0.05$).

Motor activity responses to feeding. In response to feeding, motor activity of the small intestine changed in both the milk-fed and water-fed infants compared with fasting (Table 3). Motility index increased significantly in both groups during feeding compared with fasting (Table 3; $p < 0.001$). Conversely, quiescence decreased significantly during feeding compared with fasting (Table 3; $p < 0.02$).

Timed responses of peptides and motor activity. Responses of peptides and motor activity to formula feeding were time-related. Gastrin concentration was significantly increased at 30 and 60 min ($p < 0.01$) but decreased to values similar to fasting at 150 and 180 min. GIP and PYY concentrations were significantly elevated at 30, 60, 120, and 150 min compared with fasting (all $p < 0.01$ or less). NT was significantly higher at 60 and 120 min compared with fasting but declined at 180 min to values similar to fasting. Motor activity response to feeding, characterized by the number of peaks/30 min, also changed with time. The number of peaks/30 min significantly increased compared with fasting within 30 min of the initiation of the formula infusion; they remained elevated throughout the feeding infusion and declined to fasting values within 30 min after the feeding was completed (all $p < 0.01$).

DISCUSSION

Plasma concentrations of gastrin, GIP, NT, and PYY during fasting were significantly different in preterm and term infants compared with healthy adult women. Gastrin and NT, potent stimulants of motility and peptide and acid release, were significantly lower in preterm infants compared with adults. GIP and PYY, potent inhibitors of motility and peptide secretion, were significantly higher in preterm infants compared with adults. Plasma concentrations of GIP and PYY were also significantly higher in term infants than healthy adults, suggesting that further postnatal maturation of these peptide concentrations occurs in infancy. These data concur with those showing cord blood PYY concentrations to be significantly higher in neonates than in healthy adults (8) and the presence of postnatal maturation of other peptides such as motilin and human pancreatic polypeptide (9).

All four of the peptides measured in this study are important modulators of lower esophageal sphincter tone, gastric emptying, and intestinal motility. Our previous studies have shown that small intestinal motility patterns differ in preterm infants and term infants. Furthermore, many characteristics of intestinal motor activity patterns in preterm infants "mature" with gestational age (1) and become more similar to those seen in term infants. This current study shows that gastrin and PYY significantly vary with gestational age, suggesting that there may be a temporal association of peptide concentrations and intestinal motility. Facer (10) has shown that fetal small intestine cell numbers of gastrin, GIP, and PYY increase with gestational age. We have recently shown that fasting gastrin concentration is significantly predictive of maturation of intestinal motility patterns in preterm infants (11). Therefore, plasma concentrations of these regionally distributed peptides may serve as markers in determining the functional maturation of preterm intestinal motor activity.

Both water-fed and milk-fed babies demonstrated a change in motor activity in response to enteral feedings. However, the calculation of motility index and quiescence were used to demonstrate only that a response to feeding had occurred. Although motor responses to feedings are nutrient-related in both infants (12) and adults (13), we did not characterize the motor activity responses to feeding sufficiently to determine if the responses in this study were nutrient related.

Water-fed infants demonstrated no change in plasma peptide concentration during the 180-min study. Milk-fed infants experienced a significant rise in GIP, NT, and PYY. Although babies fed by oroduodenal tube demonstrated no increase in gastrin with feeding, gastrin increased significantly in babies fed by gastric tube, suggesting that oroduodenal feedings had failed to provide adequate stimulation to antral G cells. Although G cells are present in both neonatal antrum and duodenum, the maturation of antral G cells occurs postnatally in concert with maturation of plasma gastrin concentration (14). Collectively, these data suggest that antral G cells play a more significant role than the duodenal cells in regulating neonatal gastrin response to nutrient stimulation.

The rise in peptide concentration in response to nutrients was time related. G cells are localized to the antrum and duodenum, and GIP is released in response to the presence of nutrient in the duodenum. NT is released in the jejunum and PYY in the distal ileum and colon in response to the presence of intraluminal nutrients. Therefore, the monitoring of serial peptides demonstrates indirectly that active aboral transit of intraluminal milk may have occurred. The presence of motor activity that migrates distally during fasting is thought to contribute significantly to the aboral movement of intraluminal nutrients. Many preterm infants lack this distally migrating activity. Despite this "immaturity" of preterm fasting intestinal motor activity, the postprandial rise in NT and PYY confirms that aboral movement of nutrients occurred, suggesting that neonatal nonmigrating motor activity may in fact contribute to aboral transit.

Both motor activity and peptide concentrations in preterm infants are parallel in maturity. Fasting motor activity is immature in preterm infants compared with term infants, but motor activity changes in response to feeding (2). Similarly, fasting

plasma peptide concentrations of preterm infants differ from those of adults, but peptide concentrations increase in response to feeding. The presence of this apparent functional maturity of the intestine in response to feeding suggests that the use of early enteral nutrition may be warranted in preterm infants, although the interval of feedings may need to be lengthened. The provision of early hypocaloric nutrition is associated with a rise in fasting gastrin and GIP (14). These fasting concentrations, in turn, are associated with improved fasting motor activity and clinical tolerance of routine feedings. We speculate that the monitoring of fasting motor activity and hormones are more relevant measurements of functional maturation of the preterm intestine than are feeding responses. However, the preterm intestine discerns nutrients from nonnutrients, and future studies should identify nutrients that enhance optimal functional responsiveness of the preterm intestine. These studies would provide important insights to design better feeding regimens for preterm infants.

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