Maintenance of villous height and crypt depth in piglets by providing continuous nutrition after weaning

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Abstract

Thirty-two piglets weaned at 28 days of age were used to test the hypothesis that maintenance of nutrition after weaning would prevent the normal decline in villous height and increase in crypt depth and hence preserve the structure and function of the small intestine. Piglets were allocated to one of four treatments at weaning: (1) control group killed at weaning; (2) piglets offered a dry starter diet ad libitum; (3) piglets offered ewes’ fresh milk; and (4) piglets offered ewes’ fresh milk plus 20 g L-glutamine per l. Piglets in treatments (3) and (4) were offered ewes’ fresh milk every 2 h in a feeding schedule that increased from 1-2 l per piglet on the 1st day after weaning to 2-4 l on days 4 and 5. On the 5th day all piglets were killed and samples of small intestine were taken for histological and biochemical examination. Feeding ewes’ milk or ewes’ milk plus 20 g L-glutamine per l maintained (P > 0.05) villous height and crypt depth compared with piglets killed at weaning. In contrast, piglets given a dry starter diet had shorter villi (P < 0.001), deeper crypts (P < 0.001), and proportionately 0.21 to 0.28 less protein (P > 0.05) in their intestinal mucosa. Piglets given the starter diet proportionately grew from 0.49 to 0.62 more slowly (P < 0.01), ate the same amount of dry matter (DM; P > 0.05), but consumed proportionately 0.30 less energy (P < 0.001) than their counterparts given the milk diets. No treatment differences in the specific activity of lactase and sucrase were observed (P > 0.05). Significant correlations existed between voluntary food intake and villous height at the proximal jejunum for piglets given the starter diet and ewes’ milk (P < 0.05 and P = 0.073, respectively). In turn, villous height was significantly correlated (r = 0.78 to 0.87, P < 0.05) with the rate of body-weight gain after weaning in these two groups. For piglets offered ewes’ milk plus glutamine, an increase in DM intake was associated only with increases in crypt depth (P < 0.01). These data show that the structure and function of the small intestine can be preserved when a milk diet is given after weaning, and suggest an association between food intake and villous height in determining post-weaning weight gain.

Keywords: continuous nutrition, crypt depth, glutamine, piglets, villous height.

Introduction

The small intestine of the newly weaned piglet generally undergoes a reduction in villous height and an increase in crypt depth (Gay, 1976; Gay, Barker and Moore, 1976; Hampson, 1986a and b; Miller, James, Smith and Bourne, 1986; Cera, Mahan, Cross, Reinhart and Whitmoyer, 1988; Kelly, Smyth and McCracken, 1991b and c). These changes have generally been associated with reductions in the specific activity of disaccharidase enzymes (Gay et al., 1976; Hampson and Kidder, 1986; Miller et al., 1986) and a reduced capacity of the gut to absorb xylose (Miller, Newby, Stokes and Bourne, 1984; Hampson and Kidder, 1986; Hampson and Smith, 1986) and alanine (Smith, 1984; Miller et al., 1986). The net effect of these alterations is thought to reduce the digestive and absorptive function of the gut (Gay et al., 1976; Hampson, 1983). From a production perspective these changes are important because they may reduce growth and extend the time pigs take to reach market weight.

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Numerous factors have been proposed to account for the apparent decrease in digestive and absorptive capacity of the small intestine after weaning (see Hampson, 1987; McCracken and Kelly, 1993). In many studies the possible contribution that food intake per se may play in the maintenance of gut integrity has largely been ignored, or considered not to be of major aetiological importance (e.g. Hampson, 1983), and only Kelly et al. (1991c) have examined effects of a controlled level of food intake on gut structure and function. Some workers (Robertson, Clark and Bruce, 1985; Bark, Crenshaw and Leibbrandt, 1986) have shown that after weaning there is a period of temporary starvation during which piglets fail to eat sufficient food to cover their maintenance energy requirement. McCracken (1984) and McCracken and Kelly (1984) suggested that villous atrophy and reductions in digestive enzyme activity after weaning may be related more to the lack of a continuous supply of substrate than to any antigenicity of the diet or inherently low levels of disaccharidase activity. In rodents and miniature piglets a period of food restriction (Steiner, Bourges and Leibbrandt, 1987; Altmann, 1972), or exclusion of antigenicity of the diet or inherently low levels of nutrients from the gut when animals were fed parenterally (McNeill and Hamilton, 1971; Shulman, Fiorotto, Sheng and Garza, 1984; Goldstein, Hebiguchi, Luk, Taqi, Guilarte, Franklin, Niemiec and Dudgeon, 1985; Castillo, Feng, Stevenson, Kerner and Kwong, 1990), resulted in villous atrophy and decreases in mucosal protein content and digestive enzyme activity. These findings suggest a direct role for the presence of nutrients in the lumen for the maintenance of gut structure and function after weaning.

Gut structure and function after weaning may also be limited by availability of the amino acid L-glutamine. Glutamine is the principal fuel used by the gut epithelium (Windmueller, 1982), is present in high concentrations in sow's milk around the time of weaning (Wu and Knabe, 1994), and numerous studies have demonstrated that glutamine is required by villous enterocytes to support metabolism, and the structure and function of the gut (Soubra, 1991, 1992 and 1993). Provision of oral glutamine stimulates net uptake of glutamine by enhancing brush-border transport rates (Salloum, Soubra, Fernandez and Stevens, 1990), and supports mucosal growth by stimulating the activity of glutaminase (Klimberg, Salloum, Kasper, Plumley, Dolson, Hautamaki, Mendenhall, Bova, Bland, Copeland and Soubra, 1990; Salloum, Souba, Klimberg, Plumley, Dolson, Bland and Copeland, 1989). At a time when the piglet's supply of maternal glutamine disappears, supplementation of diets with synthetic glutamine offers a means of enhancing the structure and function of the gut after weaning.

The hypothesis tested in this study was that maintenance of voluntary food intake after weaning would preserve the structure and function, and therefore maintain the digestive and absorptive capacity of the small intestine when piglets were killed 5 days later. Furthermore, we proposed that supplementation of L-glutamine in the milk diet would augment the digestive and absorptive capacity of the gut following weaning. An abstract of this work has been presented previously (Pluske, Williams and Aherne, 1991).

Material and methods

Animals and housing

Thirty-two piglets from four primiparous gilts (Large White X Landrace) bred at Medina Research Centre, Medina, Western Australia, were used in the study. Gilts were transported to the University of Western Australia on about day 75 of gestation and were housed in environmentally controlled rooms maintained at 19°C. After farrowing, litter size was standardized to eight piglets per litter. Routine management of piglets included teeth clipping, tail docking and an injection (200 mg i.m.) of iron dextran. Piglets were housed with their mothers during lactation in standard farrowing accommodation.

At an average age of 28 (s.e. 0.4) days when they weighed 8.9 (s.e. 0.34) kg, piglets were transported 300 m to an air-conditioned experimental room that contained 12 individual stainless-steel pens arranged in four banks of three pens each. Each pen had a measured floor area of 0.45 m². Ambient temperature was maintained at 25.6 (s.e. 0.44)°C with relative humidity of 0.37 (s.e. 0.07) for the 5-day duration of the study. Galvanized steel trays were placed under each pen for the collection of spilt food, faeces and urine. Two galvanised troughs were placed in each pen, one for food and the other for fresh water.

Experimental treatments

The experiment was a completely randomized block design. Piglets were allocated randomly on the basis of litter, sex and live weight to one of four treatment groups as follows: (1) sow-reared, control group killed on the day of weaning (SR) (no. = 8); (2) piglets offered a pelleted starter diet (starter) (no. = 8); (3) piglets offered ewes’ fresh milk (EM) (no. = 8); (4) piglets offered ewes’ fresh milk plus 20 g L-glutamine per 1 (EM+Gln) (no. = 8).
Table 1  Composition (g/kg) and analysis of the starter diet

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Composition (g/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat</td>
<td>276</td>
</tr>
<tr>
<td>Barley</td>
<td>200</td>
</tr>
<tr>
<td>Oat groats</td>
<td>150</td>
</tr>
<tr>
<td>Skim-milk powder</td>
<td>250</td>
</tr>
<tr>
<td>Fish meal</td>
<td>70</td>
</tr>
<tr>
<td>Vegetable oil</td>
<td>30</td>
</tr>
<tr>
<td>Limestone</td>
<td>7</td>
</tr>
<tr>
<td>Dibasic calcium phosphate</td>
<td>8</td>
</tr>
<tr>
<td>L-lysine HCl</td>
<td>2</td>
</tr>
<tr>
<td>DL-methionine</td>
<td>1</td>
</tr>
<tr>
<td>L-threonine</td>
<td>1</td>
</tr>
<tr>
<td>Vitamin and mineral pre-mix†</td>
<td>5</td>
</tr>
</tbody>
</table>

Composition

- Dry matter‡: 889
- Crude protein (g N × 6.25)‡: 213
- Ether extract‡: 128
- Ash‡: 76
- Digestible energy (MJ/kg)§: 15.0
- Total lysine§: 14
- Calcium§: 12
- Phosphorus§: 8

† Provided the following nutrients (per kg of air-dry diet): vitamins: retinol 1500 μg, cholecalciferol 20 μg, α-tocopherol 10 mg, riboflavin 2.5 mg, niacin 16 mg, pantothenic acid 10 mg, pyridoxine 2 mg, choline chloride 140 mg, cyanocobalamin 10 μg; minerals: NaCl 3.5 g, Cu 10 mg, Zn 100 mg, Mn 20 mg, Fe 60 mg, Co 0.2 mg.
‡ Determined by proximate analysis of diet.
§ Calculated from ingredients.

Diets and feeding regimen

Piglets were offered ewes' fresh milk every 2 h for 5 days in a feeding schedule that increased from 1-2 l per pig on the 1st day after weaning (i.e. 100 ml per feed) to 1-4 l on day 2, 1-8 l on day 3, and to 2-4 l of ewes' milk on days 4 and 5. In a previous study using the same genotype, Pluske and Williams (1988) determined that 25-g-old sucking piglets drank about 50 ml per suckling. Since the sow suckles 21 to 24 time per day (Pluske and Williams, 1988), it was considered that 1-2 l offered on the 1st day after weaning equated to the milk intake of these piglets prior to weaning and would ensure a reasonable rate of live-weight gain. For piglets offered ewes' milk plus glutamine, L-glutamine was added to the milk at a rate of 20 g/l milk just prior to feeding, and shaken thoroughly to ensure that the glutamine dissolved fully.

The pelleted starter diet (Table 1) was fed ad libitum so that proportionately at least 0.2 of the previous day's intake was always in front of the piglet. On some occasions a pig was observed to urinate or defecate in its food trough. When this occurred the food was removed, weighed, and an equivalent amount of fresh pellets was replaced in the feeder.

Experimental procedure

Each day at 14.00 h all piglets were weighed and, for piglets given the starter diet, any food that had spilt into the collection trays was collected and the weight recorded. For piglets given the milk diets every 2 h, the amount offered was recorded and, after 10 to 15 min, troughs were removed from each pen and any residual milk was weighed in tared containers and recorded. Between feedings all troughs were washed and scrubbed thoroughly with hot water.

Post-mortem procedure

Food was withdrawn from piglets 2 h prior to slaughter on the afternoon of the 5th day following weaning. Sow-reared piglets were removed from the sow about 1 h prior to slaughter on the day of weaning. Weaned piglets were removed from their pens and restrained on a surgical table in a dorsal position. Following the withdrawal of 5 ml blood from the heart, piglets were killed by an injection of sodium pentobarbitone. The abdomen was opened immediately, from the sternum to the pubis, and the entire gastro-intestinal tract was removed. Two segments of small intestine (= 10 cm in length), each at distances of proportionately 0.25, 0.5 and 0.75 along the gut from the gastric pylorus to the ileo-caecal valve, were clamped with haemostats. One segment at each site was filled by syringe with 5 to 10 ml ice-cold, 10% phosphate-buffered formalin (pH 7.4) for subsequent histological examination. The adjacent segment at each site was filled with an equivalent volume of ice-cold, 0.01 mol/l phosphate-buffered saline (pH 7.4), excised, wrapped immediately in aluminium foil, and then immersed in liquid nitrogen for subsequent disaccharidase and mucosal protein determinations. Total processing time from killing to obtaining gut samples was about 20 min. In the text, sites at 0.25, 0.5 and 0.75 are used interchangeably with their approximate position along the small intestine, i.e. proximal jejunum, mid jejunum and distal ileum, respectively, or are referred to as sites 1 to 3.

Histology

After fixation for several days, ring-shaped lengths of small intestine from all three sites were excised, dehydrated, and embedded in paraffin wax. From each of these, six transverse sections (4 to 6 μm) were cut, stained with haematoxylin and eosin, and examined under a light microscope. Measurements of villous height and crypt depth were taken only from sections where the plane of section ran vertically from the tip of the villus to the base of an adjacent crypt. From each section a calibrated eyepiece graticule was used to measure 10 of the tallest well oriented villi from tip to crypt mouth, and 10 associated crypts from crypt mouth to base (after Hampson, Fu and Smith, 1988).
Digestive enzyme determinations

Methods used for the determination of lactase (β-galactosidase; EC 3.2.1.23) and sucrase (sucrose-α-glucosidase; EC 3.2.1.48) activity were adapted from techniques described by Kidder and Manners (1980) and Hampson (1983). Approximately 200 mg of mucosa was gently scraped from partially thawed lengths of small intestine using a spatula. The mucosa was then placed into a plastic vial containing 40 ml of distilled water to make a 1/200 (w/v) mucosal homogenate. Following homogenization for 30 s in a polytron (Model CH-6010, Kinematica, Kriens-Luzern, Switzerland) at setting no. 10, the contents were transferred to a plastic centrifuge tube and spun at 3000 r.p.m. for 15 min. The supernatant was decanted, and 5 ml was kept in a plastic tube at 4°C for analysis of lactase and sucrase activity. A further 5 ml was retained for determination of mucosal protein concentration.

Substrate concentrations and incubation conditions used for lactase and sucrase determinations were the same as those used by Kidder and Manners (1980) and Kelly (1985). Briefly, equal quantities of substrate and 0.2 mol/l phosphate buffer were mixed thoroughly in a 50 ml volumetric flask. From this solution 200 µl was dispensed into clean glass test tubes, a further 100 µl supernatant or distilled water (blank) was added, and then all tubes were incubated in a water bath set at 37°C for exactly 30 min. After 2 min the reaction was terminated by submerging the tubes in boiling water for 2 min. The free glucose liberated by the action of lactase and sucrase was then determined using the glucose-6-phosphate dehydrogenase (EC 1.1.1.49)-hexokinase (EC 2.7.1.1) assay (Boehringer-Mannheim Biochemica, Germany) for the UV determination of glucose, as adapted by Kelly (1985).

Mucosal protein concentration

The protein content of the mucosal homogenate was determined using the method described by Gornall, Bardawill and David (1949), as adapted by Kelly (1985).

Analysis

Samples of ewes' fresh milk were collected in plastic vials and frozen at −20°C for analysis of fat, protein and total solids. Milk was analysed using a MilkoScan® (Foss Electric, Denmark) automated analyser. The proportion of fat, protein and total solids contained in ewes' milk (no. = 36) was 73 (s.e. 4.0), 48 (s.e. 1.6) and 185 (s.e. 3.5) g/kg respectively. The calculated gross energy (GE) content of the milk was 465 MJ/kg, or 25.14 MJ/kg dry matter (DM), according to the equation of Perrin (1958).

Levels of free plasma glutamine were determined according to the method of Jones and Gilligan (1983) using a Varian 5000 high-performance liquid chromatograph and a Varian Flourichrom detector. The internal standard for glutamine was 0.0913 g α-amino-guanidino-propionic acid dissolved in 100 ml double-deionized water to yield a concentration of 5 mmol/l. Plasma urea and alkaline phosphatase were measured using a biochemical analyser (COBAS MIRA, Roche Diagnostica, Switzerland).

Statistical analysis and presentation of results

All data were subjected to one-way analysis of variance for treatment effects using SYSTAT® (Wilkinson, 1990). As this experiment was conducted over two time periods owing to the availability of piglets, time was included as an independent variable in the initial analysis of variance. The effect of time was not significant for any of the variables measured, so the data were reanalysed with treatment group being the only independent variable. Pairwise comparisons between treatment means were made using Fisher's-protected least significant difference (LSD) procedure (Maindonald, 1992). Where appropriate, simple linear regressions were performed using SYSTAT® (Wilkinson, 1990).

The weight of the empty body was determined by multiplying the starting live weight of each piglet by the proportion of the carcass that was empty body in the piglets killed at weaning (Noblet and Etienne, 1987). This was 981 (s.e. 6.8) g/kg (CV = 0.0069). Empty body-weight gain was calculated, therefore, as the difference between the recorded weight of the empty body at slaughter 5 days after weaning and the estimated weight of the empty body at weaning. The protein content of the mucosa and the specific enzyme activity are presented as the mean of all three sites (0.25, 0.5 and 0.75) along the small intestine.

Results

All piglets offered the milk diets began drinking within 8 h of weaning, and became accustomed to the 2-hourly feeding schedule very quickly. Once conditioned to the procedure, piglets eagerly awaited the arrival of their food. When the trough was placed in the pen, piglets drank their milk voraciously. Most animals consumed all their milk within 1 to 3 min and there was no spillage. The health of all piglets during the experiment was excellent, and no diarrhoea was observed except on the morning of the final day when several piglets given the milk diets developed a very mild scour.
Table 2 Villous height and crypt depth of piglets killed at weaning or 5 days later

<table>
<thead>
<tr>
<th>Proportion of intestine</th>
<th>Treatment†</th>
<th>s.e.d.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>SR</td>
<td>Starter</td>
<td>EM</td>
</tr>
<tr>
<td>Villous height (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>550&lt;sup&gt;a&lt;/sup&gt;</td>
<td>330&lt;sup&gt;b&lt;/sup&gt;</td>
<td>569&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>0.50</td>
<td>464&lt;sup&gt;a&lt;/sup&gt;</td>
<td>316&lt;sup&gt;b&lt;/sup&gt;</td>
<td>428&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>0.75</td>
<td>264</td>
<td>261</td>
<td>258</td>
</tr>
<tr>
<td>Mean‡</td>
<td>426&lt;sup&gt;b&lt;/sup&gt;</td>
<td>299&lt;sup&gt;a&lt;/sup&gt;</td>
<td>418&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Crypt depth (µm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>116&lt;sup&gt;a&lt;/sup&gt;</td>
<td>182&lt;sup&gt;c&lt;/sup&gt;</td>
<td>137&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>0.50</td>
<td>124&lt;sup&gt;a&lt;/sup&gt;</td>
<td>179&lt;sup&gt;b&lt;/sup&gt;</td>
<td>137&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>0.75</td>
<td>97&lt;sup&gt;a&lt;/sup&gt;</td>
<td>172&lt;sup&gt;c&lt;/sup&gt;</td>
<td>123&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mean‡</td>
<td>112&lt;sup&gt;b&lt;/sup&gt;</td>
<td>178&lt;sup&gt;b&lt;/sup&gt;</td>
<td>132&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a,b,c</sup> Within rows, means not followed by a common superscript differ significantly.
† SR: piglets killed at weaning; Starter: piglets given dry starter diet ad libitum; EM: piglets given ewes' fresh milk; EM + Gln: piglets given ewes' fresh milk plus 20 g/1 L-glutamine.
‡ Mean of all three sites along the small intestine.

Villous height and crypt depth

Feeding ewes' milk or ewes' milk plus glutamine maintained (P > 0.05) villous height at sites 0.25 and 0.5 along the small intestine compared with piglets killed at weaning. In contrast, piglets given the starter diet had villi that were proportionately from 0.26 to 0.72 shorter (P < 0.01) at both sites than all other treatment groups. Villous height at the distal ileum was similar (P > 0.05) between treatments. Mean villous height along the length of the small intestine was proportionately 0.34 to 0.45 lower (P < 0.001) in piglets given the starter diet than it was in piglets killed either at weaning or given both milk diets. Feeding ewes' milk or ewes' milk plus glutamine did not reduce mean villous height in comparison with piglets killed at weaning. Villous height decreased (P < 0.001) from the proximal to the distal part of the small intestine (Table 2).

Crypt depth at the proximal jejunum had increased in all treatments by the time piglets were killed 5 days after weaning. This increase was less for piglets given ewes' milk or ewes' milk plus glutamine (P < 0.001) and was greatest for piglets offered the starter diet (P < 0.001). Piglets given the starter diet showed a 0.32 proportional increase in crypt depth at the proximal jejunum in comparison with both groups of piglets given milk (182 µm v. an average of 138 µm, P < 0.001). At the mid jejunum and distal ileum, crypt depth was similar in piglets killed at weaning and those given ewes' milk or ewes' milk plus glutamine (P > 0.05), but was always greater in piglets given the starter diet (P < 0.001). Mean crypt depth in piglets given ewes' milk or ewes' milk plus glutamine was, on average, 16 µm deeper (P > 0.05) than in piglets killed at weaning whereas, in piglets given the starter diet, mean crypt depth was proportionately 0.35 to 0.59 deeper than in other groups (P < 0.001). There was a trend (P = 0.128) for crypt depth to decrease from the proximal to the distal part of the small intestine (Table 2).

Piglet performance

Empty body-weight gain of piglets given the starter diet after weaning was 252 g/day. This was proportionately 0.49 less (P = 0.023) than piglets offered ewes' milk and 0.62 less (P = 0.006) than piglets given ewes' milk plus glutamine. Voluntary food intake between treatments was similar (P > 0.05), however piglets given both milk diets consumed proportionately 0.30 more energy (7.45 v. 5.7 MJ GE per day, P = 0.006) than those given the starter diet (Table 3). The variation in voluntary food intake of piglets given the starter diet was twice that of animals given the milk diets (CV: 0.25 v. 0.12).

Piglets offered ewes' milk or ewes' milk plus glutamine were more efficient in converting DM into empty body gain than piglets given the starter diet (0.8 and 0.7 v. 1.3, P < 0.001). However the inclusion of one piglet in the initial data set for starter-fed piglets, whose stomach contained = 750 g of food at slaughter but grew only 280 g after weaning, resulted in an increase in food conversion ratio (FCR) from 1.3 to 1.8. This pig was considered aberrant and was therefore removed from the calculations for FCR and energetic efficiency of body gain. The energetic cost of 1 g of empty body-weight gain was around 19 kJ digestible energy, and was similar (P > 0.05) for all treatments (Table 3).

Pattern of voluntary food intake after weaning

On the 1st day after weaning piglets eating the starter diet consumed less DM (129 v. 184 g,)
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Table 3 Performance of piglets after weaning

<table>
<thead>
<tr>
<th>Treatment†</th>
<th>SR</th>
<th>Starter</th>
<th>EM</th>
<th>EM + Gln</th>
<th>s.e.d.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Live weight (kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>weaning</td>
<td>8.6</td>
<td>8.9</td>
<td>9.1</td>
<td>8.9</td>
<td>0.87</td>
<td></td>
</tr>
<tr>
<td>after 5 days</td>
<td>10.4</td>
<td>11.3</td>
<td>11.1</td>
<td></td>
<td>0.85</td>
<td></td>
</tr>
<tr>
<td>Empty body weight (kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>weaning</td>
<td>8.5</td>
<td>8.7</td>
<td>8.9</td>
<td>8.8</td>
<td>0.85</td>
<td></td>
</tr>
<tr>
<td>after 5 days</td>
<td>10.0</td>
<td>10.8</td>
<td>10.8</td>
<td></td>
<td>0.83</td>
<td></td>
</tr>
<tr>
<td>Daily gain (g/day)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>live weight</td>
<td>307a</td>
<td>435b</td>
<td>443b</td>
<td>47.6</td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>empty body weight</td>
<td>252a</td>
<td>375b</td>
<td>407b</td>
<td>50.6</td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>Voluntary food intake (g dry matter per day)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Energy intake (MJ GE per day)</td>
<td>5.7a</td>
<td>7.4b</td>
<td>7.5b</td>
<td>0.55</td>
<td>***</td>
<td></td>
</tr>
<tr>
<td>Food conversion ratio</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(g dry matter : g EBWG)$</td>
<td>1.3</td>
<td>0.8</td>
<td>0.7</td>
<td>0.45</td>
<td>***</td>
<td></td>
</tr>
<tr>
<td>Energy cost/g EBWG‡ (kJ DE per g)</td>
<td>18.0</td>
<td>20.2</td>
<td>18.2</td>
<td>0.96</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a,b Within rows, means not followed by a common superscript differ significantly.
† See Table 2.
‡ EBWG: empty body-weight gain.

$P = 0.306$) than piglets given the milk diets. DM intake between treatment groups did not differ ($P > 0.05$) on any day after weaning. However on the 5th day piglets offered both milk diets consumed, on average, 82 g DM less ($P = 0.142$) than piglets given the pelleted diet (Figure 1).

Relationships between gut morphology, voluntary food intake and the rate of gain

Significant within-treatment relationships existed between total DM intake and villous height at the proximal jejunum in piglets given the starter diet ($r = 0.78, P = 0.039$) and ewes' milk ($r = 0.65, P = 0.073$), but were not found in animals given ewes' milk supplemented with 20 g/L glutamine. Crypt depth along the small intestine of piglets given the starter diet and ewes' milk showed no relationship with total DM intake ($P > 0.05$). However for piglets given ewes' milk plus glutamine, an increase in glutamine intake explained a significant proportion of the variation in crypt depth at the proximal ($r = 0.71, P = 0.051$) and mid ($r = 0.92, P < 0.01$) jejunum. As a consequence, DM intake was highly correlated to mean crypt depth ($r = 0.88, P = 0.004$) (Table 4).

Villous height at the proximal jejunum was significantly correlated with total empty body-weight gain in piglets given the starter diet ($r = 0.78, P = 0.024$) and ewes' milk ($r = 0.87, P = 0.005$) after weaning. In contrast, no relationships existed between these parameters in piglets offered ewes' milk plus glutamine. No significant correlations ($P > 0.05$) were recorded between crypt depth and rate of gain in any treatment groups. However at sites proportionately 0.5 ($P = 0.058$) and 0.75 ($P = 0.073$) along the gut, an increase in empty body-weight gain was associated with an increase in crypt depth for piglets given ewes' milk plus glutamine (Table 5).

Digestive enzyme activity

The mean protein content of the mucosa of the small intestine did not differ between treatments ($P > 0.05$) nor show any difference along the small intestine ($P > 0.05$) at the sites sampled. However the

![Figure 1](image-url) The daily pattern of dry matter intake (g) in piglets offered either a pelleted starter diet (O), ewes' liquid milk (A), or ewes' liquid milk fortified with 20 g/L glutamine (Q) for 5 days after weaning. Values are mean ± s.e. for eight piglets per treatment.
concentration of mucosal protein in piglets given the starter diet was from 0.21 to 0.28 lower ($P = 0.574$) than in the other groups. Similarly, the specific activity of lactase and sucrase did not differ between treatments ($P > 0.05$; Table 6). The lack of statistical significance between treatment groups was due to the large variation in protein content and digestive enzyme activity that occurred. For example, the CV for the mean specific activity of lactase in piglets given ewes' fresh milk ranged from 0.36 to 1.09, whilst that of sucrase for piglets offered the starter diet varied from 0.49 to 1.23.

The specific activity of lactase declined ($P = 0.060$) from the proximal to the distal part of the small intestine. Values decreased from 95 to 68 to 38 mmol/min per g mucosal protein at sites 1 to 3 along the small intestine. Sucrase activity, however, showed no proximal to distal decline along the gut ($P = 0.371$).

### Plasma metabolites
Levels of plasma glutamine were similar ($P > 0.05$) between dietary treatments when measured 5 days after weaning, although the level of circulating glutamine was from 0.84 to 1.15 higher ($P < 0.001$) in these groups than when piglets were killed on the day of weaning. Plasma urea was three times higher ($P < 0.001$) in piglets offered ewes' milk plus glutamine than that recorded in piglets killed either at weaning or those given a solid diet after weaning. A difference of 3.8 mmol/l urea ($P < 0.001$) existed between piglets given ewes' milk plus glutamine and

### Table 4 Correlation coefficients between total dry matter intake (g) and villous height and crypt depth (µm) when measured 5 days after weaning

<table>
<thead>
<tr>
<th>Proportion of intestine</th>
<th>Treatment†</th>
<th>Starter</th>
<th>EM</th>
<th>EM + Gln</th>
</tr>
</thead>
<tbody>
<tr>
<td>Villous height</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>0.78*</td>
<td>0.65§</td>
<td>0.10</td>
<td></td>
</tr>
<tr>
<td>0.50</td>
<td>0.36</td>
<td>0.48</td>
<td>0.20</td>
<td></td>
</tr>
<tr>
<td>0.75</td>
<td>0.14</td>
<td>0.20</td>
<td>0.51</td>
<td></td>
</tr>
<tr>
<td>Mean‡</td>
<td>0.42</td>
<td>0.65§</td>
<td>0.14</td>
<td></td>
</tr>
<tr>
<td>Crypt depth</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>0.28</td>
<td>0.58</td>
<td>0.71*</td>
<td></td>
</tr>
<tr>
<td>0.50</td>
<td>0.56</td>
<td>0.49</td>
<td>0.92**</td>
<td></td>
</tr>
<tr>
<td>0.75</td>
<td>0.37</td>
<td>0.55</td>
<td>0.53</td>
<td></td>
</tr>
<tr>
<td>Mean†</td>
<td>0.35</td>
<td>0.56</td>
<td>0.88**</td>
<td></td>
</tr>
</tbody>
</table>

† See Table 2.
‡ Mean of all three sites along the small intestine.
§ $P = 0.073$.

### Table 5 Correlation coefficients between total empty body-weight gain (g) and villous height and crypt depth (µm) when measured 5 days after weaning

<table>
<thead>
<tr>
<th>Variable</th>
<th>Proportion of intestine</th>
<th>Treatment†</th>
<th>Starter</th>
<th>EM</th>
<th>EM + Gln</th>
</tr>
</thead>
<tbody>
<tr>
<td>Villous height</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>0.78*</td>
<td>0.87**</td>
<td>0.10</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.50</td>
<td>0.68§</td>
<td>0.37</td>
<td>0.53</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.75</td>
<td>0.42</td>
<td>0.14</td>
<td>0.66</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean‡</td>
<td>0.69§</td>
<td>0.71*</td>
<td>0.36</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crypt depth</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.25</td>
<td>0.28</td>
<td>0.14</td>
<td>0.41</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.50</td>
<td>0.51</td>
<td>0.30</td>
<td>0.69§</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.75</td>
<td>0.49</td>
<td>0.45</td>
<td>0.66</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean†</td>
<td>0.44</td>
<td>0.39</td>
<td>0.63</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

† See Table 2.
‡ Mean of all three sites along the small intestine.
§ $P = 0.062$.
¶ $P = 0.058$.

### Table 6 Protein content of the mucosa and the specific activity of lactase (EC 3.2.1.23) and sucrase (EC 3.2.1.48) of piglets killed at weaning or 5 days later

<table>
<thead>
<tr>
<th>Treatment†</th>
<th>Starter</th>
<th>EM</th>
<th>EM + Gln</th>
<th>s.e.d.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mucosal protein content (mg/g mucosa)†</td>
<td>134</td>
<td>106</td>
<td>150</td>
<td>148</td>
</tr>
<tr>
<td>Lactase activity (µmol/min per g protein)‡</td>
<td>67</td>
<td>64</td>
<td>76</td>
<td>60</td>
</tr>
<tr>
<td>Sucrase activity (µmol/min per g protein)‡</td>
<td>63</td>
<td>78</td>
<td>58</td>
<td>59</td>
</tr>
</tbody>
</table>

† See Table 2.
‡ Mean of all three sites along the small intestine.

### Table 7 The concentration of metabolites in the plasma of piglets killed at weaning or 5 days later

<table>
<thead>
<tr>
<th>Treatment†</th>
<th>SR</th>
<th>Starter</th>
<th>EM</th>
<th>EM + Gln</th>
<th>s.e.d.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glutamine (µmol/l)</td>
<td></td>
<td>382ab</td>
<td>705b</td>
<td>822b</td>
<td>803b</td>
<td>95.7 **</td>
</tr>
<tr>
<td>Urea (mmol/l)</td>
<td></td>
<td>2.2ab</td>
<td>2.0b</td>
<td>2.6b</td>
<td>6.4c</td>
<td>0.48 ***</td>
</tr>
<tr>
<td>Alkaline phosphatase (U/l)</td>
<td></td>
<td>428</td>
<td>326</td>
<td>310</td>
<td>335</td>
<td>26.0</td>
</tr>
</tbody>
</table>

ab: Within rows, means not followed by a common superscript differ significantly.
† See Table 2.
those given ewes' milk alone. Urea levels in piglets given ewes' milk only was elevated, proportionately on average, 0.24 \( (P < 0.001) \) above that found in either piglets killed at weaning or those given the starter diet. There was no treatment difference \( (P > 0.05) \) in the plasma concentration of alkaline phosphatase (Table 7).

**Discussion**

* Maintenance of villous height and crypt depth after weaning

Feeding ewes' milk or ewes' milk plus 20 g/L L-glutamine to piglets every 2 h for 5 days after weaning preserved villous height and crypt depth along the entire length of the small intestine compared with piglets killed at weaning. This supports our hypothesis that maintaining nutrition after weaning at levels approximating pre-weaning intake prevents villous atrophy and an increase in crypt depth. Supplementation of ewes' fresh milk with glutamine did not enhance the structure and function of the small intestine as it was similar to that in piglets receiving ewes' milk alone. This failed to support our proposition that addition of glutamine to the diet would augment the digestive and absorptive function of the gut after weaning. However, the significant correlation observed between DM intake and crypt depth (Table 5) suggests that glutamine may be an important substrate for cell renewal in the small intestine, and will be discussed later.

The preservation of villous height and crypt depth that occurred when piglets were given milk or milk plus glutamine suggests that the balance between cell loss from the villi and cell production in the crypts was not altered after weaning. In contrast, piglets given the starter diet ad libitum had villi that were shorter and crypts deeper by the 5th day after weaning compared with piglets given both of the milk diets. An increase in crypt depth is compatible with an increase in crypt-cell production rate and an overall stimulation of cell turn-over in the small intestine (Al-Mukhtar, Polak, Bloom and Wright, 1982) that has generally been associated with a reduced digestive and absorptive capacity (Gay et al., 1976; Hampson, 1983). In the present experiment there was a marked difference in the rate of empty body-weight gain of piglets given the starter diet and those given both milk diets (252 vs. an average of 391 g/day).

Despite consuming an equivalent amount of DM, piglets given both milk diets ingested significantly more energy than those offered the solid diet. A decrease in energy intake decreases both fasting heat production (Koong, Nienaber, Pekas and Yen, 1982) and the quantity of mucosa in the small intestine (Pekas, 1986a and b). The amount of mucosa in the small intestine is highly correlated with fasting heat production of the growing pig (Pekas, 1986b). Since heat production is associated with protein synthesis (Webster, 1980 and 1981), and this is most active in the gut epithelium due to the high rate of cell turnover (Cheng and Leblond, 1974; Pekas and Wray, 1991), it is likely that an increase in energy intake will supply more substrate for mucosal growth (Pekas, 1986b). In this regard the likely contribution of the 0.3 proportional difference in energy intake to both the maintenance of gut structure and function and the disparity in growth between treatments remains unknown. This will be examined in our next experiment (Pluske, Williams and Aherne, 1996).

Although we have clearly demonstrated that mucosal integrity can be maintained after weaning we do not know the importance of milk per se. In this study we used ewes' fresh milk because of (a) its similarity in gross composition to sows' milk, and (b) we reasoned that offering weaned piglets a familiar nutritional source (i.e. milk) would help overcome the interruption to food intake that normally occurs immediately after weaning. We offered ewes' milk that was collected fresh each day and offered it to piglets in an unhomogenized and unpasteurized form. Piglets adapted readily to this diet and consumed large quantities within hours of weaning whereas piglets often take a number of days to consume similar quantities of a dry diet. However, milk is known to contain a variety of biologically active compounds (Cera, Mahan and Simmen, 1987; Jaeger, Lamar, Bottoms and Cline, 1987; Koldovsky, 1989; King and Kelly, 1990; Kelly, King, Brown and McFadyen, 1991; Kelly, King, McFadyen and Travis, 1991a; Kelly, McFadyen, King and Morgan, 1992) that are likely to influence intestinal differentiation and cell turn-over. Some workers (e.g. Hampson, 1986b) have also commented that the withdrawal of specific factor(s) associated with the consumption of sows' milk might cause decreases in villous height after weaning. Therefore it is not possible to discern whether villous height and crypt depth were attenuated by the presence of these compounds in milk or if it was attributable to the actual amount of food the piglets' consumed. Delineation of these factors forms the basis of our next experiment (Pluske et al., 1996).

* Piglet performance and relationship to gut structure

DM intakes and growth rates of piglets given all three diets were excellent. Piglets offered ewes' milk or ewes' milk plus glutamine averaged between 375 and 407 g empty body-weight gain per day, and converted milk DM to empty body-weight gain with an efficiency similar to piglets sucking the sow.
Gut morphology and nutrition in piglets

(lucas and Lodge, 1961; Noblet and Etienne, 1987). These data exemplify the rapid growth potential of the young piglet, and support impressive growth rates reported by other workers (e.g. Hodge, 1974; Williams, 1976; Harrell, Thomas and Boyd, 1993) when piglets were offered milk diets. It was apparent, however, that piglets given the starter diet were limited in their ability to ingest an equivalent amount of energy as those piglets given both milk diets despite being offered ad libitum. Some authors have attributed this phenomenon to the weaned piglet having a limited 'gut capacity' (Campbell, 1989; Cranwell and Moughan, 1989; Tybirk, 1989), although the factors influencing this are not well defined. Nevertheless, we and others (Kelly et al., 1991b and c; Pluske et al., 1996) have been unable to confirm associations between a 'reduced' digestive and absorptive capacity of the small intestine with significant decreases in the activities of lactase and sucrase or the amount of xylose absorbed, suggesting that measurement of these disaccharidases may be an inappropriate measure of digestive and absorptive function of the gut.

The reduced growth rate of piglets offered the pelleted starter diet was associated with villous shortening and an increase in crypt depth but what cannot be deduced from our data is whether or not these changes were caused by the type of food per se or the fact that piglets consumed less energy. However, a positive linear relationship between food intake and villous height was observed and, in turn, between villous height and empty body-weight gain after weaning. These relationships suggest that piglets consuming more dry food after weaning grew faster because villous height was preserved. The presence of more nutrients in the lumen of the gut is likely to influence a multitude of growth factors, biologically active peptides and gut hormones that are responsible for attenuating the structure and function of the gut.

The findings in our study provide support for the theory of 'luminal nutrition' (Gleeson, Cullen and Dowling, 1972; Williamson, 1978; Diamond and Karasov, 1983) as an explanation for both intestinal structure and the variation in intestinal morphology seen along the small intestine. For piglets offered the starter diet or ewes' milk only, villous height at the proximal jejunum responded linearly to an increase in DM (Table 4). This confirms the existence of a strong relationship between nutrition and gut structure and, because villous height at the proximal jejunum can be used as a predictor of body gain (Table 5), emphasizes the importance of food intake per se in the performance of piglets after weaning. This result is not surprising since the proximal small intestine is the major site for digestion and absorption in the gut (Kidder and Manners, 1980; Diamond and Karasov, 1983; Hampson, 1983; Puchal and Buddington, 1992). The decrease in villous height and lactase activity from the proximal to the distal part of the small intestine seen in the present study most likely parallels the decreasing gradient in nutrient concentration that occurs along the gut.

Mucosal protein content and digestive enzyme activities

The mean protein content of the mucosa in piglets killed at weaning and those given ewes' milk or ewes' milk plus glutamine is in excellent agreement with results for sow-suckled piglets reported by Kelly et al. (1991b). These workers reported a protein concentration of 149 mg/g mucosa in unweaned piglets slaughtered at 22 days of age and the average protein content of both groups given milk in our experiment was identical. Similarly, the concentration of protein in the mucosa in piglets given the solid diet was similar to that recorded by these authors on the 5th day after weaning (106 v. 113 mg protein per g mucosa), and was in the range (52 to 164 mg/g fresh mucosal scraping) reported by Kidder and Manners (1980) for weaned piglets. No statistical difference for mucosal protein content between piglets given the starter diet and milk diets was found, presumably because of the small amount of mucosa scraped (200 mg) and the large CV (0.27 to 0.86) noted between samples. It was of interest, however, that the proportional reduction in mucosal protein concentration in piglets given the pelleted diet was 0.29. This is identical to the 0.3 proportional difference in energy intake between piglets given the starter diet and those offered ewes' milk and ewes' milk plus glutamine.

The mean values for specific lactase and sucrase activity observed in this experiment are lower than those recorded by Kelly et al. (1991b and c). For example, mean specific lactase activity in piglets given the starter diet on the 5th day after weaning was 64 µmol/min per g mucosal protein whereas Kelly et al. (1991b) recorded a value of 109.5 µmol/min per g mucosal protein. Similarly, the mean specific activity of sucrase was 78 as opposed to 96.2 µmol/min per g mucosal protein found by Kelly et al. (1991b). The higher activities recorded by Kelly et al. (1991b) are best explained by the younger piglets used in that experiment. Kelly et al. (1991b) weaned piglets at 14 days of age whereas weaned animals at 28 days of age. The mean villous height of piglets used in this experiment was lower by the 5th day after weaning (302 v. 424 µm) and, because lactase is located more apically on the villus (Dahlqvist and Nordström, 1966; Nordström and Dahlqvist, 1973; Kelly et al., 1991a) and lactase activity declines with age (Bailey, Kitts and Wood, 1956; Kidder and Manners, 1980; James, Smith, Tivey.
and Wilson, 1987; Sangild, Cranwell, Sørensen, Mortensen, Norén, Wetteberg and Sjöström, 1991), piglets of a younger age having longer villi would be expected to show greater lactase activity.

A salient aspect of this experiment was the poor association between in vitro estimates of lactase and sucrase activity and the preservation of villous height and crypt depth, and the high rate of body gain observed, in piglets given the milk diets. For instance, piglets consuming the dry starter diet after weaning suffered a 0.41 proportional decrease in mean villous height and 0.36 proportional increase in mean crypt depth compared with piglets given milk, yet showed no reduction in specific activities of lactase and sucrase. A reduction in villous height and increase in crypt depth is indicative of increased rates of cell production in the crypts and enterocyte migration, thereby leaving fewer mature enterocytes on the villus available for digestion and absorption (Rey, Schmutz, Rey and Jos, 1971; Smith, 1984; Hampson, 1986a). Under these circumstances we would have predicted a decrease in the activity of these enzymes. However, we caution that only the specific activity of these enzymes was measured. Expression of enzyme activity on a total basis may have yielded more meaningful data, although data presented by Kelly et al. (1991b) suggests that even this in vitro measure does not adequately reflect the large differences seen in intestinal architecture.

**Structure and function of the small intestine in piglets offered L-glutamine**

The digestive and absorptive capacity of the small intestine, as assessed by villous height and crypt depth, the specific activities of lactase and sucrase, and the rate of empty body-weight gain, was similar between piglets offered ewes' milk and those given ewes' milk supplemented with 20 g/l L-glutamine. These findings failed to support our hypothesis that glutamine would augment the structure and function of the small intestine after weaning. The most probable reason for the lack of an effect of glutamine was that piglets did not undergo starvation and/or there was no stimulation of the adrenal cortex. During these states glutamine may become a 'conditionally essential nutrient' (Souba, 1991 and 1992) for the mucosa because high rates of glutamine metabolism provide adequate amounts of key intermediates for nucleic acid biosynthesis (Klimberg et al., 1990) and mucosal renewal (Souba, 1993). Under these circumstances enterocytes would be expected to increase their demand for glutamine because it is more important than glucose as an oxidative fuel (Windmueller and Spaeth, 1980).

In the present experiment there was no period of fasting after weaning. In view of the high level of milk (and glutamine) intake achieved, the exogenous supply of glutamine was most likely in excess of that required by the epithelium. Support for this notion comes from the 1.46 proportional increase in the concentration of plasma urea (Table 7) in piglets given ewes' milk plus glutamine compared with piglets offered ewes' milk only. An increase in urea levels following glutamine supplementation was also reported by Déchelotte, Darmann, Rongier, Hecketsweiler, Rigal and Desjeux (1991). An increase in urea concentration is consistent with the hydrolysis of glutamine to glutamate by glutaminase with its subsequent deamination to ammonia and release into the portal system. Ammonia, in turn, is detoxified in the urea cycle in the liver and released as urea into the circulation.

Despite glutamine having no apparent effect on the structure and function of the small intestine after weaning, there are two aspects of this experiment that warrant further examination. First, the lack of any correlation between villous height and food intake in piglets given ewes' milk plus glutamine (Table 4) suggests that this amino acid might have had an ameliorative rôle on intestinal structure that was not entirely related to the level of food intake per se. This may occur by stimulating the activity of mitochondrial glutaminase and/or the rate of transport across enterocytes already present on the villus. Glutamine has been shown to expedite both of these processes (Klimberg et al., 1990). Secondly, the linear increase in crypt depth as glutamine intake increased is indicative of an increase in glutamine metabolism in the mucosa. An increase in glutamine transport across the enterocytes increases the activity of mitochondrial glutaminase (Klimberg et al., 1990; Salloum et al., 1990) that would, in turn, accelerate the hydrolysis of glutamine into products that can directly enter the tricarboxylic acid cycle and generate adenosine triphosphate. This energy could be used to support cell division in the proliferative zone in the crypts of Lieberkühn (Newsholme, Crabtree and Ardawi, 1985; Klimberg et al., 1990) since glutamine is a requisite substrate for de novo DNA biosynthesis.

Several recent studies in the weaned piglet provide some support to these speculations. Wu and Knabe (1993 and 1994) have highlighted the possible importance of glutamine as an energy substrate for the small intestine of the pig both during suckling and after weaning. Wu and Knabe (1994) reported that glutamine was the most abundant free and protein-bound amino acid in sow's milk at days 22 and 29 of lactation. After weaning, Wu and Knabe (1993) measured glutamine metabolism in isolated enterocytes and found two-and 10-fold increases in the rate of oxidation of glutamine to CO₂ in
enterocytes from 29-day-old weaned piglets compared with 21-day-old sucking piglets. These data suggest that glutamine may serve as an increasingly important energy substrate for the enterocytes of weaned piglets. Rates of glutamine oxidation were not measured in the present study, although tentative support for the findings of Wu and Knabe (1993) comes from the significant linear relationship observed between glutamine intake and crypt depth (Table 4). However to observe a beneficial effect of supplemental glutamine on the digestive and absorptive capacity of the small intestine and therefore on growth rate, it is likely that an experiment lasting longer than 5 days is required. This would give cells that are formed in the crypt base time to migrate up the villus and develop full digestive and absorptive capacity. In this regard, Meier, Knabe, Wu and Borbolla (1993) reported that the addition of 10 g/kg glutamine to a maize-soya-bean diet prevented villous atrophy in the jejenum on the 7th day after weaning.

We have demonstrated that villous height and crypt depth can be maintained if piglets do not suffer the nutritional stress of interrupted intake immediately after weaning. In contrast piglets offered a solid, pelleted diet at the same level of voluntary food intake suffered a decrease in villous height and an increase in crypt depth after weaning. Piglets drinking ewes’ milk and ewes’ milk plus glutamine, however, ingested proportionately 0.30 more energy than animals eating the solid diet. The possible contribution of this increase to the preservation of gut structure and function and the disparity in body-weight gain could not be resolved from this study, but will be examined in the next experiment (Pluske et al., 1996).

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