Effects of Moist Extruded Full-fat Soybeans on Gut Morphology and Mucosal Cell Turnover Time of Weanling Pigs

Shiyan Qiao, Defa Li*, Jianyang Jiang[1], Hongjie Zhou[2], Jingsu Li[2] and P. A. Thacker[3]
Ministry of Agriculture Feed Industry Center, China Agricultural University, Beijing, P. R. China

ABSTRACT : Ten barrows, weaned at 28 days (7.2±0.1 kg BW), were used to evaluate the effects of feeding extruded full-fat soybeans on intestinal morphology and mucosal cell turnover time. All pigs were fed corn-based diets with half of the pigs receiving diets supplemented with 15.5% soybean meal and 3% soybean oil and the remaining pigs fed a diet in which the soybean meal and oil were replaced by 18.5% extruded full-fat soybeans. The pigs were individually placed in 80×150 cm metabolic cages and fed twice daily an amount approximately equal to their *ad libitum* intake for a period of 14 days. On day 14, pigs were weighed and then injected intraperitoneally with [^{3}H]thymidine (100 μCi/kg of BW, specific activity 20 Ci/mmol) 6 h after the morning meal. A pig from each treatment was killed 1, 4, 8, 16, or 24 h post-injection and intestinal tissues were collected. Daily gains for pigs fed the soybean diet and extruded full-fat soybean diet were 0.24 and 0.31 kg/day ($p=0.05$) with feed conversions of 1.58 and 1.39 ($p=0.05$), respectively. In comparison with pigs fed soybean meal, pigs fed moist extruded full-fat soybeans had a decreased crypt depth in their duodenum and cecum ($p<0.1$), while the villus height in the mid jejunum and ileum and the total height (villus height plus crypt depth) of the ileum and mid jejunum increased ($p<0.05$). The villus width in the duodenum and mid jejunum decreased ($p<0.05$). The number of crypt epithelial cells in the upper jejunum increased but decreased in the ileum, colon and cecum ($p<0.05$). The number of villus epithelial cells in the ileum and the upper and mid jejunum increased ($p<0.05$). The time for migration of epithelial cells in the crypt-villus column decreased ($p<0.05$) in all sites except the upper jejunum, ileum and cecum. The mucosal turnover rate for all intestinal sites except the upper jejunum, colon and cecum decreased ($p<0.05$). From these data, we conclude that inclusion of moist extruded full-fat soybeans in weanling pig diets can improve the intestinal morphology and slow the migration rate and turnover time of epithelial cells of the small intestine, especially in the mid jejunum compared with soybean meal. ***(Asian-Aust. J. Anim. Sci. 2003. Vol 16, No. 1 : 63-69)***

Key Words : Extruded Soybeans, Intestinal Morphology, Cell Turnover Time, Weanling Pigs

INTRODUCTION

Solvent extracted soybean meal is the most common source of supplementary protein used in swine production (Swick, 1994). Its popularity can be attributed to several factors including its widespread availability, its high protein, lysine and energy content as well as its palatability (De Schutter and Morris, 1990). Unfortunately, soybean meal is poorly utilized by young pigs (Dunsford et al., 1989; Li et al., 1991; Friesen et al., 1993). The reduction in performance has been attributed to a transient hypersensitivity to soybean meal (Newby et al., 1984) in which villus height decreases and crypt depth increases due to a cell-mediated immune response in the small intestine (Li et al., 1991).

Extruded full-fat soybeans are a potential alternative to soybean meal for use in weaned pig diets. The inclusion of extruded full-fat soybeans has been shown to improve growth performance and increase nutrient digestibility for weanling pigs (Geurin et al., 1988; Friesen et al., 1993; Woodworth et al., 2001). The mechanism by which this improvement is achieved has not been fully elucidated. In this study, weanling pigs were used to investigate the influence of moist extruded full-fat soybeans on pig performance and cell morphology in the intestine. In addition, migration time and turnover rates of intestinal epithelial cells were measured to understand more about this process.

* Address reprint request to Defa Li. Tel: +86-10-62893588, Fax: +86-10-62893688, E-mail: defali@public2.bta.net.cn
[1] Laiyang Agricultural College, Laiyang, China.
[2] Institute of Atomic Energy, China Academy of Agriculture Science, Beijing, China.
[3] Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Canada.
[4] This project was sponsored by grants from the China Natural Science Foundation of China.
Received March 5, 2002; Accepted July 22, 2002

MATERIALS AND METHODS

Animals and diets

Ten crossbred barrows (Duroc × Landrace × Yorkshire) weaned at 28 days (7.2±0.1 kg BW) were fed corn-based diets with half of the pigs receiving diets supplemented with 15.5% soybean meal and 3% soybean oil and the remaining pigs receiving a diet in which the soybean meal and oil were replaced by 18.5% extruded full-fat soybeans obtained from the China Agriculture Machine Academy (Table 1). The barrows were placed in 80×150 cm metabolic cages in an environmentally controlled nursery (26°C) and fed twice daily (08:00 and 18:00 h) an amount approximately equal to their *ad libitum* intake for 14 days. Both diets were fed in mash form. Water was available at all times. On day 0 and day 14, the pigs were weighed and average daily gain, feed intake and

Table 1. Ingredient composition and chemical analysis of the experimental diets

	Soybean meal[1] +oil	Extruded full fat soybean[2]
Ingredients (% as fed)		
Corn	64.96	65.00
Soybean meal	15.50	-
Soybean oil	3.0	-
Extruded full fat soybeans	-	18.5
Plasma protein	5.0	5.0
Whey powder	3.0	3.0
Fish meal	4.0	4.0
Dicalcium phosphate	2.1	2.1
Lime stone	0.85	0.85
Salt	0.3	0.3
Vitamin-mineral premix[3]	1.0	1.0
L-Lysine	0.23	0.19
DL-Methionine	0.06	0.06
Chemical analysis (% as fed)		
Crude protein	19.09	19.01
Calcium	0.97	0.99
Phosphorus	0.79	0.82
Lysine	1.22	1.24
Methionine	0.35	0.36

[1] Analyzed crude protein and urease activity were 43.4% and 0.08 change in pH.

[2] Analyzed crude protein, urease activity, content of trypsin inhibitor (TIU), protein solubility were 35.49%, 0.04 pH, 2.6 TIU/mg and 75.89% respectively.

[3] Contributed per kilogram of diet: vitamin A, 5512 IU; vitamin D_3, 551 IU; vitamin E, 66.1 IU; riboflavin, 5.5 mg; vitamin K_3, 2.2 mg; vitamin B_{12}, 27.6 μg; pantothenic acid, 14 mg; niacin, 30.3 mg; choline, 551 mg; Fe, 100 mg; Cu, 120 mg; Mn, 100 mg; Zn, 100 mg; Se, 0.3 mg; I, 0.3 mg.

feed conversion were calculated for the 14 d treatment period. On day 14, pigs were fed normally. Six hours postprandially, all pigs (10 in total) were weighed and [^{3}H]thymidine (Specific activity 20 Ci/mmol, Institute of Isotope, Chinese Atomic Energy Academy) was injected (100 μCi/kg of BW) intraperitoneally. The [^{3}H]thymidine was diluted with sterile saline to 100 μCi/mL before injection.

Tissue collection

One pig from each treatment was killed at either 1, 4, 8, 16 or 24 h after the [^{3}H]thymidine injection. Pigs were euthanized with excess halothane. Their gastrointestinal tracts were quickly removed, and divided into the duodenum, upper jejunum, mid jejunum, lower jejunum, ileum, cecum and colon. The duodenum was defined as that portion of the intestine from the pylorus to the point where the bile duct entered the intestinal tract (approximately 10 cm). The ileal sample was collected 10 cm away from the end point of the ileum, which was differentiated by the ileal-cecal valve and the ileal-colonic junction. Jejunum samples were taken from the upper (upper jejunum), mid (mid jejunum) and lower (lower jejunum) portion of this section of the intestine between the duodenum and the ileum. The cecum and colon samples were taken from the middle of the cecum and spiral colon respectively. Sections 4-6 cm in length were taken from the middle of the above intestinal sites, gently flushed with saline, ligated at both ends with string, and filled with 10% buffered formalin using a syringe. These tissues were placed into 10% buffered formalin.

Morphology and autoradiography

Formalin fixed tissue was embedded into paraffin, and sliced into 7-micron sections. They were mounted on glass slides, the paraffin were removed with xylene and then hydrated with increasing proportions of water in ethanol. After hydration, the slides were coated with NTB-2 emulsion (Kodak, Japan) using the method of Kopriwa and Leblond (1962). Slides were stored at 4°C for a 14-25 d exposure period and then developed. After developing, the slides were stained with hematoxylin and eosin.

The structure of the gut regions was observed using an Olypus BX-50 Microscope. The numbers of epithelial cells in the crypt and villus column were counted with an eye-piece micrometer at a magnification of 400×, and the villus height, crypt depth, and villus width were measured at a magnification of 100×. The cell size was calculated using the height of the column divided by the number of cells in the column. The distance travelled by the leading labelled cells was determined at a magnification of 400×. All measurements were made from the basement membrane at the bottom of the crypt to the tip of the villi, and only continuous cell columns on unbranched villi were measured (Imondi and Bird, 1966). Columns were defined as a single row of epithelial cells extending from the base of the crypts to the tip of the villi. Ten columns were counted for each gut region of each pig, and these counts were averaged (Radecki et al., 1992).

The turnover time of the epithelial cells was calculated as the amount of time necessary for the leading labelled cells in the crypt or villi to reach the tip of the crypt (cecum and colon) or villi (small intestine) (Imondi and Bird, 1966). Plots were made with the percentage of the distance from the bottom of the crypts to the tip of the villi covered by the leading labelled cells in different gut regions at various intervals after [^{3}H]thymidine injection, and regression equations presented as $y=a+bx$ were obtained. In these regression equations, b (slope of the regression line) is the migration rate of the labelled cells. Each gut site was plotted. Figure 1 is the plot of the mid jejunum. Time for mucosal turnover was estimated from the migration rates (Radecki et al., 1992).

Chemical analysis

Samples of all feeds were analyzed for their nitrogen, calcium and total phosphorus content using the methods of the AOAC (1990). Nitrogen was analyzed using the Kjeldahl method (AOAC official method 988.05), calcium by titration

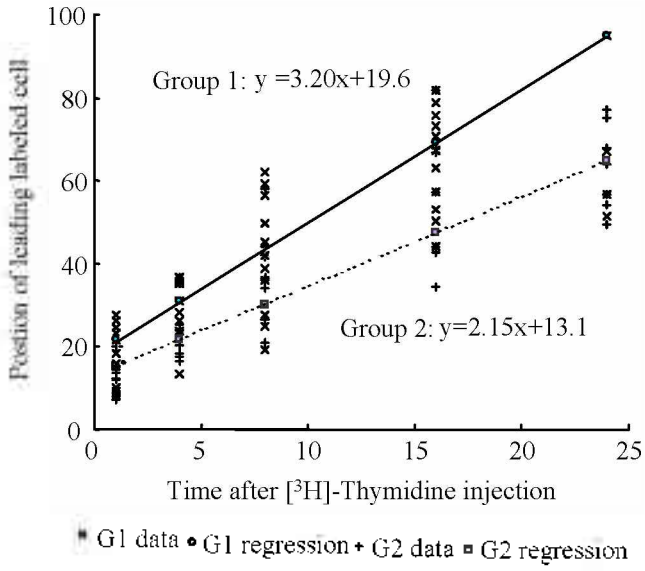

Figure 1. Epithelial cell migration of mid jejunum along the crypt-villus columns. Group 1 (G1) represented the soybean meal diet, r=0.94. Group 2 (G2) represented the extruded full-fat soybean diet, r=0.90.

with 0.1 N $KMnO_4$ (AOAC official method 927.02) and the total phosphorus was determined colorimetrically using a molybodovanadate reagent (AOAC official method 965.17).

Lysine and methionine content in feed were determined using high-performance liquid chromatography (Shimadzu LC 10 Liquid Chromagraph, Kyoto, Japan). Urease activity in samples of soybean meal and extruded full-fat soybeans were analyzed using the method of Yangsheng (1993). The sample of extruded full-fat soybeans was hydrolyzed with 0.2% KOH, the protein solubility was analyzed using the method of Yangsheng (1993). The content of trypsin inhibitor was determined with an enzyme-chemical method (Kakade et al., 1974).

Statistical analysis

Average daily gain, feed intake and feed conversion, as well as morphology including crypt depth, villus height, villus width, the number and size of the epithelial cells along crypt-villus column of gastrointestinal tract were analyzed as a one-way analysis of variance (SAS, 1999), with pig as the experimental unit. Time for mucosal turnover was estimated using linear regression equations determined for each pig at various sites in the gastrointestinal tract, regressing the location of the labeled cell on the crypt-villus column over time.

RESULTS

Pigs fed extruded full-fat soybean diet had higher weight gain and feed conversion (p=0.05) on d 14 than those fed the soybean meal plus soybean oil diet, but dietary inclusion of extruded full-fat soybeans did not modify feed intake (Table 2). The villi of pigs fed extruded full-fat soybean diet tended to be more slender than the villi of pigs fed the soybean meal plus soybean oil diet (Figure 2 and Figure 3). The villi of pigs fed the soybean meal diet were often flattened and fused to one another (Figure 2). In addition, malformed villi in the small intestinal mucosa were found in pigs fed the soybean meal diet. Subjective

Table 2. Effect of feeding soybean meal or extruded full-fat soybeans on the performance of weanling pigs

	Soybean meal+oil	Extruded full fat soybean	SEM[1]	P value
Average daily gain (kg)	0.24[a]	0.31[b]	0.02	0.05
Average daily feed intake (kg)	0.38	0.43	0.03	0.46
Feed conversion	1.58[a]	1.39[b]	0.06	0.05

[ab] Means in the same row followed by different superscripts differ at the p values indicated.
[1] Standard error of the mean.

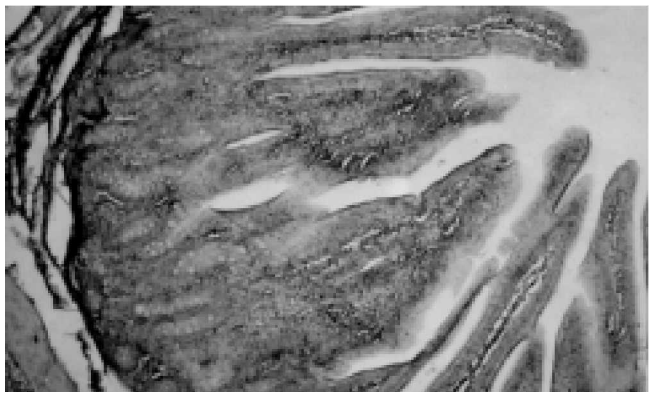

Figure 2. Jejunum crypts and the center of the villus in pigs fed soybean diet were invaded by lymphocytes and plasma cells. Figure showed deeper crypts and expanded villus with some of the villus stuck together (100×magnification).

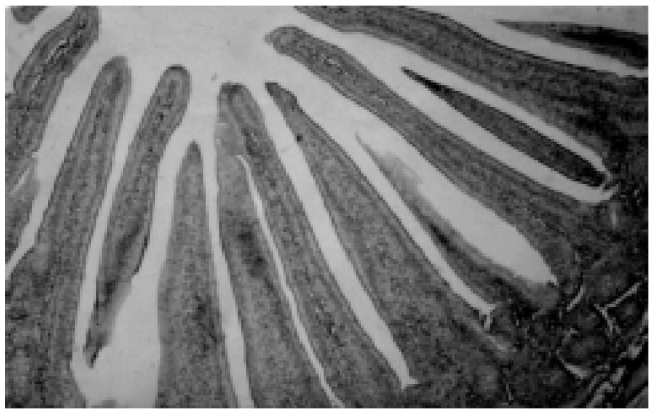

Figure 3. The structure of the jejunum crypts and villus of a pig fed the extruded full-fat soybean diet. Figure showed shallow crypts, with long and slender villus.

observations (as seen under the microscope) indicated a higher concentration of lymphocytes and plasma cells in the lamina propria of pigs fed the soybean meal diet than pigs fed the extruded full-fat soybean diet.

Dietary treatment influenced crypt depth, villus height, and villus width quite dramatically, especially at the mid jejunum (Table 3). Compared with pigs fed the soybean meal diet, pigs fed the extruded full-fat soybean diet had a lower crypt depth in their duodenum (p=0.07) and cecum (p=0.08), a higher villus height, and total height (crypt depth plus villus height) in their mid jejunum (p=0.02) and ileum (p=0.02), and a lower villus width in the duodenum (p=0.04) and mid jejunum (p=0.004).

Table 3. Effect of feeding soybean meal or extruded full-fat soybeans on the crypt depth and villus height of intestinal mucosa of weanling pigs[1, 2]

Intestinal site	Soybean meal +oil	Extruded full-fat soybean	SEM[3]	P value
Crypt depth (μm)				
Duodenum	467[a]	396[b]	30	0.07
Upper jejunum	334	287	42	0.32
Mid jejunum	288	265	23	0.34
Lower jejunum	295	331	24	0.18
Ileum	265	303	22	0.13
Colon	473	427	28	0.15
Cecum	451[a]	401[b]	24	0.08
Villus height (μm)				
Duodenum	758	687	106	0.54
Upper jejunum	697	767	65	0.32
Mid jejunum	543[a]	661[b]	36	0.02
Lower jejunum	496	500	100	0.97
Ileum	394[a]	503[b]	39	0.03
Total height (μm)				
Duodenum	1,225	1,083	127	0.32
Upper jejunum	1,031	1,054	89	0.80
Mid jejunum	831[a]	926[b]	31	0.02
Lower jejunum	791	831	80	0.63
Ileum	659[a]	806[b]	49	0.02
Villus width (μm)				
Duodenum	178[a]	132[b]	16	0.04
Upper jejunum	136	116	12	0.15
Mid jejunum	147[a]	113[b]	8	0.004
Lower jejunum	133	116	14	0.30
Ileum	138	127	7	0.14

[a,b] Means in the same row followed by the different superscripts differ at the p values indicated

[1] Each mean represents 5 pigs, with ten crypt-villus columns per pig.

[2] No villus occur in the colon and cecum.

[3] Standard error of the mean.

The effects of dietary treatment on the number of epithelial cells along the crypt-villus column are shown in Table 4. Compared with pigs fed the soybean meal diet, pigs fed the extruded full-fat soybean diet had a less number of crypt epithelial cells in the ileum (p=0.05), colon (p=0.03) and cecum (p=0.01), but a greater number in the upper jejunum (p=0.04). Pigs fed the extruded full-fat soybean diet had significantly greater number of villus epithelial cells in their upper jejunum (p=0.04), mid jejunum (p=0.004) and ileum (p=0.09).

The dietary treatments also influenced the crypt cell size (Table 5). The crypt cell size of pigs fed the extruded full-fat soybean diet was significantly greater in the ileum (p=0.03) and cecum, but significantly smaller in the upper jejunum (p=0.03). Dietary treatment did not influence the villus cell size at any intestinal sites.

Diet had a significant effect on both migration rate and turnover rate of epithelial cells (Table 6). Pigs fed the extruded full-fat soybean diet had a lower migration rate (um/h) for epithelial cells in the duodenum (p=0.01), mid jejunum (p=0.01) and lower jejunum (p=0.01) than pigs fed the soybean meal diet, but had a higher migration rate in the upper jejunum (p=0.02) and cecum (p=0.01).

Compared with the soybean meal diet, extruded full-fat soybean diet slowed the mucosal turnover rates in the duodenum (p=0.04), mid jejunum (p=0.01) and ileum (p=0.02), but speeded rates in the cecum (p=0.02) and colon (p=0.02). There was no difference in intestinal mucosal turnover rate in the upper jejunum and lower jejunum between the two dietary treatments.

DISCUSSION

Previous studies (Guerin et al., 1988; Friesen et al., 1993) have demonstrated that moist extruded soy products can be included in a high-nutrient-dense early-weaned pig diet. Terlington et al. (1990) reported that pigs fed moist extruded soy protein concentrate gained more from day 0 to day 14 than pigs fed a milk diet. Li et al. (1991) stated that pigs fed moist-extruded soy protein concentrate and those fed milk protein diet had similar weight gains. Friesen et al. (1993) compared raw soy flakes, commercial soy protein concentrate and extruded commercial soy protein concentrate in 21 day weanling pig diets, and indicated that pigs fed extruded commercial soy protein concentrate had higher weight gains and feed conversion (feed:gain), suggesting that the increased gains resulted from a higher feed intake. Our experiment showed pigs fed moist extruded full-fat soybean diet gained more (p=0.05) and had a higher feed conversion (p=0.05) than those fed the

Table 4. Influence of extruded full-fat soybeans on the number of epithelial cells in the intestinal mucosa[1,2]

Intestinal site	Soybean meal+oil	Extruded full-fat soybean	SEM[3]	P value
Number of epithelial cells in crypt				
Duodenum	58	66	13	0.56
Upper jejunum	39[a]	58[b]	7	0.04
Mid jejunum	58	60	15	0.92
Lower jejunum	51	52	7	0.80
Ileum	57[a]	45[b]	5	0.05
Colon	82[a]	72[b]	3	0.03
Cecum	85[a]	64[b]	4	0.01
Number of epithelial cells in villus				
Duodenum	100	120	11	0.11
Upper jejunum	114[a]	181[b]	16	0.04
Mid jejunum	107[a]	151[b]	11	0.004
Lower jejunum	96	115	18	0.33
Ileum	86[a]	107[b]	10	0.09
Total number of epithelial cells in crypt and villus				
Duodenum	158	186	16	0.13
Upper jejunum	153[a]	239[b]	13	0.000
Mid jejunum	165[a]	211[b]	21	0.05
Lower jejunum	147	167	13	0.16
Ileum	143	152	19	0.34

[a,b] Means in the same row followed by the different superscripts differ at the p values indicated.
[1] Each mean represents 5 pigs, with ten crypt-villus columns per pig.
[2] No villus occur in the colon and cecum.
[3] Standard error of the mean.

Table 5. Effect of extruded full-fat soybeans on the epithelial cell size of intestinal mucosa[1,2]

Intestinal site	Soybean meal+oil	Extruded full-fat soybean	SEM[3]	P value
Crypt cell size (μm)				
Duodenum	8.5	6.5	1.51	0.23
Upper jejunum	8.7[a]	5.1[b]	1.20	0.03
Mid jejunum	5.8	4.6	1.09	0.31
Lower jejunum	6.0	6.4	0.51	0.53
Ileum	4.7[a]	6.8[b]	0.67	0.04
Colon	5.8	5.9	0.61	0.78
Cecum	5.3[a]	7.8[b]	0.65	0.01
Villus cell size (μm)				
Duodenum	7.7	5.8	1.41	0.24
Upper jejunum	6.3	4.3	0.64	0.30
Mid jejunum	5.2	4.4	0.42	0.11
Lower jejunum	5.1	4.4	0.44	0.13
Ileum	4.5	4.8	0.54	0.60

[a,b] Means in the same row followed by the different superscripts differ at the p values indicated.
[1] Each mean represents 5 pigs, averaged from 50 observations.
[2] No villus occur in the colon and cecum.
[3] Standard error of the mean.

commercial soybean meal diet, indicating that moist extruded full-fat soybeans is a suitable feed ingredient for weanling pigs.

In addition to enhanced feed intake, it is likely that the extrusion process reduced the concentration of anti-nutritional factors in the diet. For example, Friesen et al. (1993) reported decreased trypsin inhibitor concentration as

Table 6. Influence of extruded full fat soybeans on the turnover rate of the gastrointestinal tract mucosa in weanling pigs[1,2]

Intestinal site	Soybean meal +oil	Extruded full-fat soybean	SEM[3]	P value
Migration rate (μm/h)				
Duodenum	29.6[a]	21.6[b]	0.79	0.000
Upper jejunum	22.1[a]	25.3[b]	0.47	0.02
Mid jejunum	21.9[a]	15.9[b]	0.44	0.000
Lower jejunum	21.7[a]	17.3[b]	0.36	0.000
Ileum	18.6[a]	20.0[b]	0.48	0.07
Colon	8.8	8.5	0.49	0.30
Cecum	7.5[a]	9.1[b]	0.23	0.000
Migration rate (cells/h)				
Duodenum	4.7[a]	4.2[b]	0.14	0.02
Upper jejunum	4.4[a]	7.1[b]	0.10	0.000
Mid jejunum	4.9[a]	4.5[b]	0.12	0.04
Lower jejunum	4.9[a]	4.2[b]	0.08	0.02
Ileum	4.8	4.6	0.12	0.12
Colon	1.7	1.7	0.09	0.84
Cecum	1.7	1.7	0.12	0.37
Turnover rate (h)				
Duodenum	28.3[a]	34.7[b]	1.84	0.04
Upper jejunum	32.6	33.5	1.15	0.65
Mid jejunum	25.1[a]	40.4[b]	1.49	0.000
Lower jejunum	28.6	31.8	1.10	0.15
Ileum	24.8[a]	31.1[b]	1.46	0.02
Colon	44.0[a]	36.8[b]	1.55	0.02
Cecum	42.4[a]	34.0[b]	1.79	0.02

[a,b] Means in the same row followed by different superscripts differ at the p values indicated.
[1] Each mean represents 5 pigs, averaged from 50 observations.
[2] Standard error of the mean.

a result of moist extrusion of various soybean products. Such an effect would increase the level of amino acids available for protein synthesis in the body.

Source of the protein used in weanling diets has been shown to cause changes in the morphology of the small intestine (Miller et al., 1984a,b). Villus heights were generally greater for pigs fed hydrolysed casein than for pigs fed soybean meal, and pigs fed soybean meal had greater ($p<0.05$) lumina propria depth at all intestinal locations (Dunsford et al., 1989). Dunsford et al. (1989) also found the villi of pigs fed soybean meal were wider than that of pigs fed hydrolysed casein. They observed that the villi of pigs fed soybean meal fused to one another, and there was a higher concentration of lymphocytes and plasma cells in the crypt of pigs fed soybean meal, indicating the immunological responses occurring in the small intestinal wall for pigs fed soybean meal were more active than for pigs fed hydrolysed casein. Newby et al. (1984) suggested that a transient hypersensitivity response to dietary antigens could be the predisposing factor causing the morphological changes.

Values of villus height and crypt depth determined in this experiment were within the range of previously reported values (Moon, 1970; Li et al., 1990; 1991). Li et al. (1991) reported that pigs fed moist extruded commercial concentrate soybean protein diet had a higher villus height and a lower crypt depth than pigs fed soybean meal diet. Intestinal damage resulting from soybean meal can be quantified by the amount of xylose absorbed into the blood stream, and decreased xylose absorption corresponds to decreased villus height and increased crypt depth caused by the hypersensitivity response from antigen existed in the soybean meal (Li et al., 1990). In our study, pigs fed moist extruded full-fat soybean diet had higher villus height and a lower crypt depth than pigs fed soybean meal diet.

Radecki et al. (1992) reported that the number of epithelial cells in the crypt-villus column ranged from 49.0 in the colon to 98.2 in the jejunum, and the size of epithelial cells ranged from 6.4 μm in the jejunum and ileum to 7.2 μm in the colon. In our study, the number of epithelial cells in the crypt-villus column ranged from 64 in the cecum to 211 in the mid jejunum, the size ranged from 4.23 μm in the mid jejunum to 7.99 μm in the duodenum. Pigs fed the extruded full-fat soybean diet had a greater number and a smaller size of small intestinal mucosal cells than pigs fed the soybean meal diet. This means the villus of pigs fed the extruded full-fat soybean diet had a greater absorptive area than that of pigs fed the soybean meal diet.

This experiment provides evidence that mucosal epithelial cells migrate from the crypts to the villus tips. For example, 8 h following [^{3}H]thymidine injection, the leading labeled cell appeared in the crypt-villus conjunction of the jejunum (Figure 4). Quastler and Sherman (1959) reported that it took about nine hours for labelled cells to appear on the villi in rats. Imondi and Bird (1966) noted that labelled cells appeared on the villi eight hours after injection for chicks.

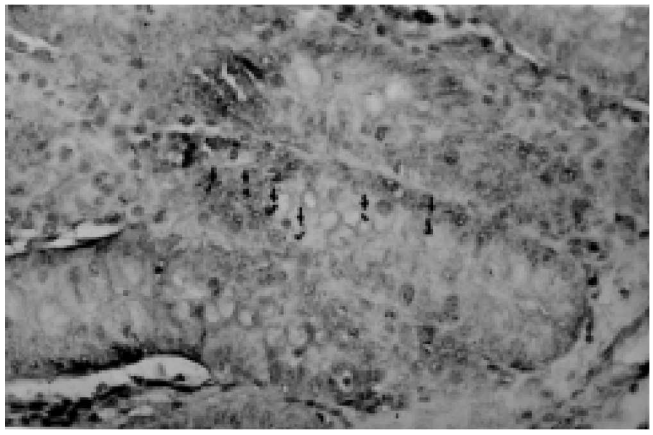

Figure 4. Mid jejunal photo-microradioautograph 8 h following [^{3}H]thymidine injection. The leading labelled cells (indicated by the arrows) were approaching the crypt-villus conjunction.

Only a very few papers have reported cell turnover rate of pigs. Age has a great effect on the turnover rate of intestinal mucosa, and different intestinal sites have different turnover rates. Moon (1970) reported that as pig's age, the turnover rate of intestinal mucosa decreased. For a one day old pig, the turnover rate of small intestinal mucosa was 7-10 d, but for a 21 day old pig, this time decreased to 2-4 d. Radecki et al. (1992) reported that the turnover rate of the intestinal mucosa of a 35 day old pig was from 33.8 h in the lower jejunum to 101.6 h in the ileum. In our study, the turnover rates were lower than those reported by Moon (1970) and Radecki et al. (1992).

The maintenance energy requirements of the gastrointestinal tract have been estimated to be approximately 24% of the total daily maintenance energy requirements of animals (Yen et al., 1988). Radecki et al. (1992) suggested that a decrease in turnover rate of mucosal cells may decrease the maintenance energy requirements of the gastrointestinal tract. In our study, the turnover rate of the duodenum, mid jejunum and ileum of pigs fed extruded full-fat soybean diet decreased significantly ($p<0.01$ or 0.05), and the turnover rate of the lower jejunum tended to be slower than pigs fed soybean meal diet. Thus, extruded full-fat soybeans in the weanling diet may decrease the maintenance energy requirements of the small intestine. The key factor causing the slower turnover rate of the small intestinal mucosa is that pigs fed extruded full-fat soybean diet had a greater crypt-villus height.

IMPLICATIONS

From the results of this experiment, it seems that using full-fat soybeans produced by a moist extruder to replace

soybean meal plus soybean oil in post-weanling diets can improve piglet performance and intestinal mucosal morphology, including crypt depth, villus height and villus width. Also, extruded full-fat soybeans in weanling pig diets can slow the replacement rate of small intestinal epithelial cells, reducing the amount of energy required to maintain the small intestine.

REFERENCES

AOAC. 1990. Official Methods of Analysis. 15th edn. Association of Official Analytical Chemists, Arlington, Virginia.

De Schutter, A. C. and J. R. Morris. 1990. Soybeans: Full-fat. In: Non-traditional Feed Sources for Use in Swine Production (Ed. P. A. Thacker and R. N. Kirkwood). Butterworths, Stoneham, Massachusetts. pp. 439-451.

Dunsford, B. R., D. A. Knabe and W. E. Hanesly. 1989. Effect of dietary soybean meal on the microscopic anatomy of the small intestine in the early weaned pigs. J. Anim. Sci. 67:1855-1863.

Friesen, K. G., J. L. Nelssen, R. D. Goodband, K. C. Behnke and L. J. Kats. 1993. The effect of moist extrusion of soy products on growth performance and nutrient utilization in the early-weaned pigs. J. Anim. Sci. 71:2099-2109.

Geurin, H. B., G. A. Kesel, W. T. Black, T. B. Hatfield and C. N. Daniels. 1988. Effect of isolated soy protein and whey on replacing dried skim milk in a prestarter for weaned baby pigs. J. Anim. Sci. 66 (Suppl. 1):320 (Abstr.).

Imondi, A. R. and F. H. Bird. 1966. The turnover of intestinal epithelium in the chick. Poult. Sci. 45:142-147.

Kakade, M. L., J. J. Rackis, J. E. McGhee and G. Puski. 1974. Determination of trypsin inhibitor activity of soy products: A collaborative analysis of an improved procedure. Cereal Chem. 51:376-382.

Kopriwa, B. and C. P. Leblond. 1962. Improvements in the coating technique radioautography. J. Histochem. 10:269-284.

Li, D. F., J. L. Nelssen, P. G. Reddy, F. Blecha, J. D. Hancock, G. L. Allee, R. D. Goodband and R. D. Klemn. 1990. Transient hypersensitivity to soybean meal in the early weaned pig. J. Anim. Sci. 68:1790-1799.

Li, D. F., J. L. Nelssen, P. G. Reddy, F. Blecha, R. D. Klemn and R. D. Goodband. 1991. Interrelationship between hypersensitivity to soybean proteins and growth performance in early-weaned pigs. J. Anim. Sci. 69:4062-4069.

Miller, B. G., T. J. Newby, C. R. Stokes, D. J. Hampson, P. J. Brown and F. J. Bourne. 1984a. The importance of dietary antigen in the cause of post-weaning diarrhoea in pigs. Am. J. Vet. Res. 45:1730-1733.

Miller, B. G., T. J. Newby, C. R. Stokes and F. J. Bourne. 1984b. Influence of diet on post-weaning malabsorption and diarrhoea in the pig. Res. Vet. Sci. 36:187-193

Moon, H. W. 1970. Epithelial cell migration in the alimentary mucosa of the suckling pig. Proc. Soc. Exp. Biol. Med. 137:151-156.

Newby, T. J., B. G. Miller, C. R. Stokes, D. J. Hampson and F. J. Bourne. 1984. Local hypersensitivity response to dietary antigens in early weaned pigs. In: Recent Advances in Animal Nutrition (Ed. W. Haresign and D. J. A. Cole). Butterworths, London. pp. 49-59.

Quastler, H. and F. G. Sherman. 1959. Cell population kinetics in the intestinal epithelium of the mouse. Exp. Cell Res. 17:420-438.

Radecki, S. V., P. K. Ku, M. R. Bennink, M. T. Yokoyama and E. R. Miller. 1992. Effect of dietary copper on intestinal mucosa enzyme activity, morphology, and turnover rates in weanling pigs. J. Anim. Sci. 70:1424-1431.

SAS. 1999. SAS Users Guide, Version 8, SAS Institute Inc., Cary, NC.

Swick, R. A. 1994. Soybean meal quality. American Soybean Association Technical Bulletin PO11-1994.

Terlington, W. H., D. S. Pollmann, J. G. Endres and J. A. Coalson. 1990. Soy protein concentrates: An alternative to milk proteins. Feedstuffs, September, p. 16.

Woodworth, J. C., M. D. Tokach, R. D. Goodband, J. L. Nelssen, P. R. O'Quinn, D. A. Knabe and N. W. Said. 2001. Apparent ileal digestibility of amino acids and digestible and metabolizable energy content of dry extruded-expelled soybean meal and its effects on growth performance of pigs. J. Anim. Sci. 79:1280-1287.

Yangsheng, S. 1993. Methods of Feed Analysis and Feed Quality Determination. Beijing Agricultural University Press, Beijing, China, pp. 260-267.

Yen, J. T., J. A. Nienaber, D. A. Hill, and W. G. Pond. 1988. Oxygen consumption by whole body and by portal drained organs in swine. J. Anim. Sci. 66(Suppl. 1):331(Abstr.).