Effect of dietary protein source on feed intake, growth, pancreatic enzyme activities and jejunal morphology in newly-weaned piglets

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Seventy piglets with no access to creep feed were weaned at 28 d of age and fed on one of four diets based on either skimmed-milk powder (SMP), soya-bean-protein concentrate (SPC), soya-bean meal (SBM) or fish meal (FM). At 0, 3, 6 and 10 d after weaning, piglets were killed and the pancreas and digesta from stomach and small intestine were collected, freeze-dried and analysed for dry matter (DM), N, and trypsin (*EC* 3.4.21.4) and chymotrypsin (*EC* 3.4.21.1) activities. Small-intestinal tissue samples were taken to examine gut wall morphology. Results indicated that dietary protein source affected post-weaning feed intake, pancreatic weight, gastric pH and gastric protein breakdown, and pancreatic and jejunal trypsin and chymotrypsin activities. Post-weaning feed intake appeared to be an important factor in digestive development of newly-weaned piglets.

Pancreas: Dietary protein: Piglet

In pigs, digestive disorders are frequently encountered in the early post-weaning period. The protein digestive capacity of newborn piglets is adapted to the digestion of milk proteins. Proteins of plant origin are digested to a lesser extent than milk proteins resulting in poor performance when fed to newly-weaned piglets (Wilson & Leibholz, 1981*a*, *c*). It has been hypothesized (Jones, 1986) that this may be due to insufficient development of the pancreatic enzyme system, since many authors report a decline in enzyme activities following weaning (Lindemann *et al.* 1986; Owsley *et al.* 1986). However, the often impaired feed intake of newly-weaned piglets may also play a role in this respect.

The present experiment was designed to study the development of pancreatic enzymes of newly-weaned piglets in relation to dietary protein source and post-weaning feed intake.

MATERIALS AND METHODS

Seventy piglets with no access to creep feed during the suckling period were weaned at 28 d of age. Ten piglets were anaesthetized immediately after weaning (day 0), weighed and exsanguinated from the jugular veins and artery. During exsanguination the gastrointestinal tract was divided into four segments: stomach, duodenum (first 2 m from pylorus), ileum (last 2 m of the small intestine) and jejunum (remaining part of the small intestine). Digesta were collected quantitatively from the stomach and small intestine segments. Usually, death of the piglets occurred after the tissue and digesta samples had been collected. In fresh gastric digesta trichloroacetic acid (TCA)-precipitable protein was measured according to Ternouth *et al.* (1974). Digesta samples were weighed, the pH was measured and samples were frozen and stored at −20° before freeze-drying. The pancreas was excised, freed from adhering tissues, frozen and stored at −20° before freeze-drying. After freeze-drying, stomach and jejunum digesta samples were ground (1 mm) and analysed for dry matter and crude protein (N × 6·25) content.

The TCA-precipitable protein fraction of the digesta comprises proteins and large peptides which cannot be absorbed by the intestinal wall without further hydrolysis (Souffrant, 1991). The ratio between TCA-precipitable protein (PP) and crude protein (CP) in gastric digesta was therefore calculated to determine the degree of gastric protein breakdown.

Trypsin (*EC* 3.4.21.4) and chymotrypsin (*EC* 3.4.21.1) activities were measured in jejunal digesta and in pancreatic tissue after freeze-drying (Bergmeyer, 1974; Van Baak *et al.* 1991).

The remaining sixty piglets were fed on one of four experimental diets based on either skimmed-milk powder (SMP), soya-bean-protein concentrate (SPC), soya-bean meal (SBM) or fish meal (FM). Diet compositions are given in Table 1. At 3, 6 and 10 d after weaning, five piglets per diet were anaesthetized 1 h after feeding 100 g diet. Samples were collected, processed and analysed as described above (twenty piglets/d). At 6 d after weaning, duplicate tissue samples (approximately 5 mm × 5 mm) were taken from the proximal and distal jejunum immediately after removing the small intestine from the body cavity. These tissue samples were stored in 3 ml cryotubes (Sanbio BV Biological Products, P.O. Box 540, NL-5400 AM, Uden The Netherlands), frozen immediately in liquid N and kept at −70° until further analysis. Sections were cut from the deep-frozen tissue samples within 2 weeks from collection using a cryostat (2800 Frigocut N; Reichert-Jung, Heidelberg, Germany), stained with toluidine blue and the lengths of ten villi and the depths of ten crypts were measured in each sample.

Analysis of variance was performed using the GLM procedure of SAS (Statistical Analysis Systems, 1990) for each day of sampling separately using the following model:

$$Y_{ij} = \mu + D_i + (b_1 \times \text{FI}) + e_{ij},$$

in which Y_{ij} is the dependent variable, μ is the overall mean, D_i is the effect of diet ($i = 1, 2, 3, 4$), $b_1 \times \text{FI}$ is the effect of feed intake (overall co-variable) and e_{ij} is the error term.

Initially the effect of feed intake within diet was also tested, but this was found to be not significant for all variables except for chymotrypsin activity 3 d after weaning and was therefore eliminated from the model. Since the overall effect of feed intake on chymotrypsin activity on day 3 was not significant, the effects of diet and of feed intake within diet on chymotrypsin activities at day 3 were analysed according to the following model:

$$Y_{ij} = \mu + D_i + (b_{1i} \times \text{FI}) + e_{ij},$$

in which Y_{ij} is the dependent variable (chymotrypsin activity at day 3), μ is the overall mean, D_i is the effect of diet ($i = 1, 2, 3, 4$), $b_{1i} \times \text{FI}$ is the effect of feed intake within diet and e_{ij} is the error term. From analysis of the data on feed intake during the first 3 d after weaning, two distinct patterns of feed intake emerged: several piglets consumed less than 50 g feed during these 3 d, while the others ingested more than 100 g. These two groups of piglets were called 'non-eaters' and 'eaters' respectively, and differences between these groups were analysed statistically. Post-hoc analysis of the effects of feed intake and diet on enzyme activities at day 3 was performed with the 'eaters' only.

To evaluate the development of gastrointestinal tissue weights after weaning the following model was used:

$$Y_{ij} = \mu + W_i + e_{ij},$$

Table 1. *Composition of experimental diets (g/kg)*

Diet...	SMP	SPC	SBM	FM
Ingredient				
Skimmed-milk powder*	470·0	—	—	—
Soya-bean-protein concentrate†	—	254·0	—	—
Soya-bean meal‡	—	—	344·0	—
Fish meal§	—	—	—	213·0
Maize/wheat starch	295·0	289·0	207·8	341·7
Dextrose	133·4	109·6	109·6	109·6
Lactose	—	225·0	225·0	225·0
Soya-bean oil	20·0	22·0	27·5	20·0
Cellulose	50·0	41·0	28·5	50·0
Ground limestone	7·5	14·5	14·5	7·5
$CaHPO_4$	5·0	22·5	21·0	2·5
NaCl	3·0	3·0	3·0	3·0
$KHCO_3$	3·0	2·0	—	11·0
$NaHCO_3$	1·0	2·0	4·0	4·0
L-Lysine HCl	—	1·4	1·6	—
DL-Methionine	1·1	2·2	2·1	1·0
L-Threonine	—	0·8	0·4	0·7
Vitamin and mineral premix	10·0	10·0	10·0	10·0
Cr_2O_3	1·0	1·0	1·0	1·0
Calculated contents				
Dry matter	938·0	939·5	933·0	938·8
Crude protein (N × 6·25)	162·1	164·4	163·8	162·2
Ether extract	25·1	24·4	31·8	35·4
Crude fibre	49·5	49·5	50·2	49·5
Inorganic matter	54·6	57·9	59·9	50·5
Net energy (MJ/kg)	10·20	10·25	10·21	10·49
Analysed contents				
Dry matter	912·1	924·1	927·0	925·3
Crude protein (N × 6·25)	163·9	169·7	145·5	148·4
Buffering capacity‖	7·2	6·4	6·4	7·7
Pellet hardness¶	14·25	18·00	3·88	4·38

SMP, skimmed-milk powder; SPC, soya-bean-protein concentrate; SBM, soya-bean meal; FM, fish meal.
* Crude protein, 351 g/kg; trypsin inhibitor activity, < 0·5 mg inhibited trypsin/g product; antigens, 2 titre $\log_2$; protein dispersability index, 0·93.
† Soycomil P, Loders Croklaan BV, P.O. Box 4, 1520 AA Wormerveer, The Netherlands. Crude protein, 639 g/kg; trypsin inhibitor activity, 1·3 mg inhibited trypsin/g product; antigens, < 1 titre $\log_2$; protein dispersability index, 0·04. Heat treated.
‡ Crude protein, 392 g/kg; trypsin inhibitor activity; 1·6 mg inhibited trypsin/g product; antigens, 5 titre $\log_2$; protein dispersability index, 0·12. Heat treated.
§ Crude protein, 692 g/kg; trypsin inhibitor activity; 1·1 mg inhibited trypsin/g product; antigens, 2 titre $\log_2$; protein dispersability index, 0·10.
‖ Volume (ml) 1 M-HCl needed to reach pH 4·00 in a suspension of 20 g feed in 100 ml demineralized water.
¶ Determined by Kahl Pellet Tester (Amandus Kahl Nachf. Maschinenfabrik, 2057 Reinbek, Hamburg). Pellet hardness expressed in kgf.

in which Y_{ij} is the dependent variable (gastric or intestinal tissue weight), W_i is the day after weaning ($i = 0, 3, 6, 10$) and e_{ij} is the error term.

RESULTS

Feed intake and growth

At weaning piglets were 28 d of age and had a mean live weight of 7·1 kg (SD 1·3, n 70). Feed intake during the first 3 d post-weaning was affected by dietary protein source (Table 2).

Table 2. *Effect of diets containing skimmed-milk powder (SMP), soya-bean-protein concentrate (SPC), soya-bean meal (SBM) or fish meal (FM) on average daily feed intake (FI) and average daily growth (ADG) of newly-weaned piglets**

(Values are least square means)

Day...		0–3	3–6	6–10
FI(g/d)				
n		60	40	20
SMP		56[a]	286	354
SPC		80[ab]	318	406
SBM		95[b]	314	376
FM		112[b]	339	453
	SEM	14	23	39
ADG (g/d)				
n		60	40	20
SMP		26	224	268
SPC		−6	293	257
SBM		−0	215	173
FM		26	273	217
	SEM	27	45	47
Effect of feed intake:				
Linear regression coefficient		2·56	0·27	0·43
P (regression coefficient = 0)		$P < 0.001$	NS	NS

NS, not significant.

a, b Least square means within a column without a common superscript were significantly different ($P < 0.05$).

* For details of diets and procedures, see Table 1 and pp. 354–355.

Pellets of the SMP diet were harder than pellets of the SBM and FM diets (Table 1) and feed intake on the SMP diet was lower during the first 3 d post-weaning. Feed intake on the SPC diet was intermediate although pellets of this diet were very resistant to crumbling. Ten piglets out of sixty (five on SMP, three on SPC and two on SBM) consumed less than 50 g feed during the first 3 d after weaning. From 3 d after weaning, feed intake was similar for all diets. Average daily gain was not affected by dietary protein source (Table 2). During the first 3 d after weaning, growth was very variable between piglets and feed intake strongly affected average daily gain. From 3 d after weaning, no effect of feed intake on growth was found (Table 2).

Tissue weights

Dietary protein source did not influence the weight of the stomach and the small intestine (absolute and relative to live weight) after weaning (Table 3). The relative weight of the small-intestinal tissue decreased during 3 d after weaning (Fig. 1). The relative weight of gastric tissue increased gradually after weaning (Fig. 1).

Pancreatic weight was clearly affected by dietary protein source. At day 3 the weight of the pancreas (g/kg live weight) was higher for piglets fed on SPC and FM than for piglets fed on SBM. At day 6 the pancreatic weight (g) was high for piglets fed on SPC and SMP and low for piglets fed on FM. At day 10 the relative weight of the pancreas was high for piglets fed on SPC and low for piglets on the FM diet (Table 3). At days 6 and 10 post-weaning, small-intestinal tissue weight was positively affected by preceding feed intake. At day 6 the same was found for the weight of the pancreas (absolute and relative).

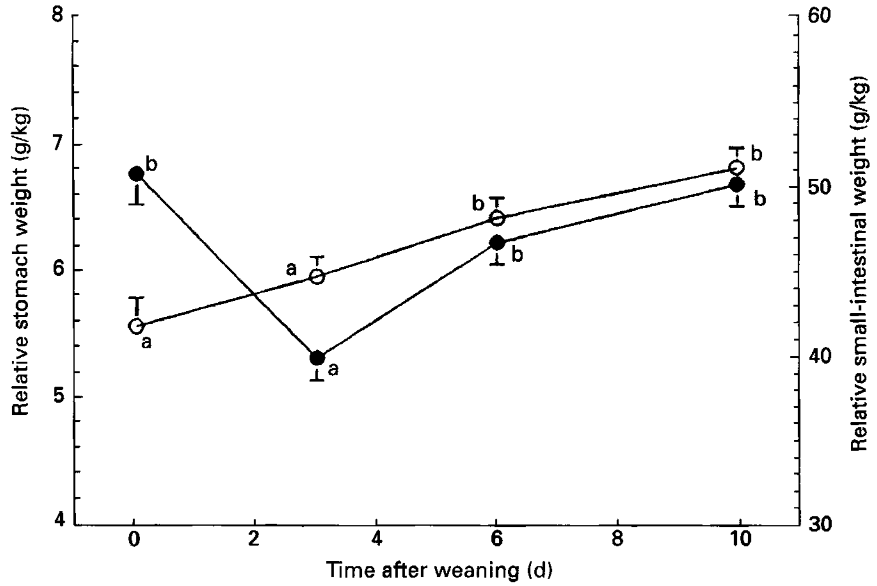

Fig. 1. Development of relative stomach (○) and small intestinal (●) tissue weights (g/kg live weight) after weaning. Values are least square means, with their standard errors indicated by vertical bars. Data points within a tissue carrying different letters were significantly different ($P < 0{\cdot}05$).

Digesta pH

At days 3 and 6 after weaning, digesta pH was not affected by dietary protein source. At day 3, jejunum pH was positively related to feed intake (Table 4). At day 10, piglets fed on the soya-bean diets had lower gastric pH than piglets fed on the FM diet.

Gastric PP:CP ratio

At day 10 after weaning the PP:CP ratio in the stomach digesta was significantly lower for piglets fed on the FM diet than for piglets on SPC, indicating a higher degree of gastric protein breakdown in gastric digesta at the time of slaughter with piglets fed on the FM diet (Table 4).

Gut wall morphology

None of the piglets killed at day 6 post-weaning had finger-shaped villi. All villi were tongue- and leaf-shaped (Kik *et al.* 1990). No effects of dietary protein source on villus lengths or crypt depths were found. Jejunal villus lengths (proximal and distal) and distal crypt depths at day 6 after weaning were positively related to feed intake (Table 4).

Enzyme activities

Enzyme activities are presented in Table 5 (trypsin) and in Table 6 (chymotrypsin).

Four out of twenty piglets sampled at day 3 post-weaning had consumed less than 10 g feed daily and these piglets were analysed separately because they ('non-eaters') were distinctly different from the other piglets with respect to enzyme activities. Non-eaters had higher enzyme activities in pancreatic tissue and higher chymotrypsin activities/g jejunal digesta than piglets that did consume appreciable amounts of feed after weaning ('eaters'; Table 7). Trypsin activity:chymotrypsin activity in pancreas and jejunum was higher for 'eaters' than for 'non-eaters' (Table 7). Therefore, the 'non-eaters' were not included in the further statistical analysis of piglets sampled at day 3 after weaning (Tables 5 and 6).

Table 3. *Effect of diet and feed intake on digestive organ tissue weights (g and g/kg live weight (LW)) and digesta weights (g) of newly-weaned piglets**

(Least square means and standard errors of the mean)

	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*†	*P*
Day 0										
Tissue wt (g)										
Stomach	38·2	3·13								
Small intestine	345·5	21·42								
Pancreas	10·37	0·843								
Tissue wt (g/kg LW)										
Stomach	5·6	0·23								
Small intestine	51·0	1·85								
Pancreas	1·53	0·081								
Digesta wt (g)										
Stomach	113·9	26·62								
Small intestine	124·8	24·86								
Day 3										
Tissue wt (g)										
Stomach			42·7	41·8	43·3	43·2	NS	4·18	0·023	NS
Small intestine			300·7	263·6	313·1	279·6	NS	26·54	0·336	NS
Pancreas			11·39	11·46	10·02	10·73	NS	0·881	−0·006	NS
Tissue wt (g/kg LW)										
Stomach			6·2	5·9	5·5	6·3	NS	0·29	−0·001	NS
Small intestine			39·7	38·4	39·9	41·8	NS	2·98	0·003	NS
Pancreas			1·53[ab]	1·66[b]	1·27[a]	1·59[b]	0·019	0·084	−0·003	0·012
Digesta wt (g)										
Stomach			177·1	136·6	130·2	129·9	NS	29·57	1·354	0·002
Small intestine			94·7	71·2	124·1	79·2	NS	19·12	0·797	0·003
Day 6										
Tissue wt (g)										
Stomach			54·5	54·4	48·7	47·2	NS	3·34	0·057	0·063
Small intestine			369·9	404·8	362·1	354·8	NS	23·55	0·657	0·005
Pancreas			13·47[bc]	15·41[c]	11·68[ab]	10·91[a]	0·003	0·717	0·038	< 0·001

Table 3. (*cont.*)

	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*†	*P*
Tissue wt (g/kg LW)										
Stomach			6·6	6·2	6·7	6·3	NS	0·29	−0·003	NS
Small intestine			44·7	46·1	49·8	47·0	NS	2·30	0·009	NS
Pancreas			1·63	1·76	1·64	1·41	NS	0·109	0·002	0·019
Digesta wt (g)										
Stomach			125·0	204·7	202·1	191·8	NS	32·25	−0·127	NS
Small intestine			158·0	258·2	213·8	167·5	NS	37·14	−0·049	NS
Day 10										
Tissue wt (g)										
Stomach			62·9	57·3	64·7	60·3	NS	6·03	0·073	NS
Small intestine			480·6	438·4	459·0	421·3	NS	31·04	0·924	0·006
Pancreas			15·62	17·59	14·52	13·17	NS	1·314	0·019	NS
Tissue wt (g/kg LW)										
Stomach			6·7	6·5	7·4	7·0	NS	0·36	−0·002	NS
Small intestine			51·6	49·9	51·7	48·6	NS	2·18	0·037	0·087
Pancreas			1·67[abc]	1·98[c]	1·65[ab]	1·54[a]	0·044	0·108	−0·000	NS
Digesta wt (g)										
Stomach			198·7	166·4	273·5	202·0	NS	40·79	0·737	0·072
Small intestine			207·5	185·5	188·4	129·5	NS	39·29	−0·169	NS

LSM, least square mean; SMP, skimmed-milk powder; SPC, soya-bean-protein concentrate; SBM, soya-bean meal; FM, fish meal; NS, not significant.

[a, b, c] Least square means within a row without a common superscript were significantly different ($P < 0·05$).

* For details of diets and procedures, see Table 1 and pp. 354–355.

† Coefficient of regression.

Table 4. *Effect of diet and feed intake on digesta pH, precipitable protein:crude protein (PP:CP) ratio in the stomach, and gut wall morphology of newly-weaned piglets**

(Least square means and standard errors of the mean)

	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*†	*P*
Day 0										
pH										
Stomach	3·80	0·459								
Duodenum	5·82	0·179								
Jejunum	6·85	0·146								
Ileum	7·47	0·124								
PP:CP										
Stomach	0·64	0·043								
Day 3										
pH										
Stomach			6·38	4·99	4·64	6·40	NS	0·699	0·012	NS
Duodenum			6·49	6·10	5·90	6·50	NS	0·217	−0·000	NS
Jejunum			7·10	7·57	6·78	7·28	NS	0·204	−0·007	0·010
Ileum			7·37	7·83	7·65	7·78	NS	0·237	−0·003	NS
PP:CP										
Stomach			0·19	0·28	0·18	0·19	NS	0·056	−0·001	NS
Day 6										
pH										
Stomach			4·29	5·18	5·12	6·11	NS	0·610	−0·001	NS
Duodenum			5·98	5·76	5·94	6·22	NS	0·340	−0·002	NS
Jejunum			7·20	7·20	7·36	7·30	NS	0·168	−0·000	NS
Ileum			7·71	7·52	7·84	7·87	NS	0·148	0·001	NS
PP:CP										
Stomach			0·17	0·35	0·25	0·29	NS	0·056	0·000	NS
Villus length (μm)										
Proximal			314	301	286	296	NS	17·6	0·400	0·017
Distal			300	280	300	269	NS	18·3	0·436	0·013
Crypt depth (μm)										
Proximal			132	139	129	137	NS	5·3	0·043	NS
Distal			134	130	144	132	NS	7·9	0·148	0·043
Day 10										
pH										
Stomach			5·36[ab]	3·89[a]	4·94[a]	6·63[b]	0·017	0·539	0·004	NS
Duodenum			5·94	5·91	6·08	6·37	NS	0·261	−0·001	NS
Jejunum			7·48	7·75	7·70	7·02	NS	0·207	−0·000	NS
Ileum			8·10	7·96	7·89	8·12	NS	0·196	−0·000	NS
PP:CP										
Stomach			0·40[ab]	0·57[b]	0·42[ab]	0·27[a]	0·027	0·063	0·001	NS

LSM, least square mean; SMP, skimmed-milk powder; SPC, soya-bean-protein concentrate; SBM, soya-bean meal; FM, fish meal; NS, not significant.

[a, b, c] Least square means within a row without a common superscript were significantly different ($P < 0·05$).

* For details of diets and procedures, see Table 1 and pp. 353–355.

† Coefficient of regression.

Table 5. *Effect of diet and feed intake on trypsin activities (units*) in digesta and pancreas of newly-weaned piglets†*

(Least square means and standard errors of the mean)

	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*‡	*P*
Day 0										
Ptryp	923	394·4								
Jtryp	180	67·2								
Ptryptot	2193	1349·5								
Jtryptot	979	761·9								
tottryp	3172	1395·0								
Pt/c	1·33	0·489								
Jt/c	1·59	0·551								
Day 3 ('eaters')										
Ptryp			2025	1068	904	1287	NS	264·4	12·64	0·041
Jtryp			602[c]	360[b]	154[a]	152[a]	0·001	53·5	0·57	NS
Ptryptot			5266	2348	1974	3325	NS	863·9	31·78	0·097
Jtryptot			4149[b]	1524[a]	1011[a]	954[a]	0·001	305·8	5·76	NS
tottryp			9415[b]	3871[a]	2985[a]	4279[a]	0·015	933·7	37·54	0·074
Pt/c			3·80	3·71	2·29	2·22	NS	0·618	0·01	NS
Jt/c			5·99	4·64	3·13	3·45	NS	0·615	0·01	NS
Day 6										
Ptryp			2526	2209	2041	1718	NS	387·2	8·45	0·021
Jtryp			790[b]	649[ab]	534[ab]	382[a]	0·039	90·8	1·47	0·075
Ptryptot			7287[b]	7803[b]	5033[ab]	3832[a]	0·048	1010·2	40·40	< 0·001
Jtryptot			4285	6269	4265	3784	NS	743·6	19·38	0·008
tottryp			11571[bc]	14072[c]	9298[ab]	7615[a]	0·001	906·9	59·79	< 0·001
Pt/c			5·94	4·44	4·52	6·36	NS	0·757	0·00	NS
Jt/c			5·18	7·61	5·52	7·98	NS	0·803	0·01	NS
Day 10										
Ptryp			3407	2939	2669	4846	NS	603·5	14·20	0·024
Jtryp			837	823	734	544	NS	84·7	−0·99	0·228
Ptryptot			11327	11665	8912	14435	NS	2130·8	60·10	0·009
Jtryptot			8430	7787	7503	3746	NS	1742·5	2·48	0·881
tottryp			19757	19452	16415	18180	NS	2323·2	62·58	0·011
Pt/c			6·77	5·44	6·70	6·15	NS	0·866	0·01	0·412
Jt/c			7·00	8·29	6·48	6·42	NS	0·709	−0·00	0·807

LSM, least square mean; SMP, skimmed-milk powder; SPC, soya-bean-protein concentrate; SBM, soya-bean meal; FM, fish meal; Ptryp, trypsin activity/g freeze-dried pancreatic tissue; Jtryp, trypsin activity/g freeze-dried jejunal digesta; Ptryptot, total trypsin activity in pancreas; Jtryptot, total trypsin activity in jejunum; tottryp total trypsin activity in pancreas+jejunum; Pt/c, Ptryp/chymotrypsin activity/g freeze-dried pancreatic tissue; Jt/c, Jtryp/chymotrypsin activity/g freeze-dried jejunal digesta; NS, not significant.

[a, b, c] Least significant means within a row without a common superscript were significantly different ($P < 0.05$).

* 1 unit = amount of enzyme required to hydrolyse 1 μmol substrate (Nα-p-toluolsulphonyl-L-arginine methyl ester)/min at 25° and pH 8·1.

† For details of diets and procedures, see Table 1 and pp. 353–355.

‡ Coefficient of regression.

Table 6. *Effect of diet and feed intake on chymotrypsin activities (units*) in digesta and pancreas of newly-weaned piglets*†

(Least square means and standard errors of the mean)

	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*‡	*P*
Day 0										
Pchym	796	188·9								
Jchym	105	14·2								
Pchymtot	1899	634·1								
Jchymtot	560	103·2								
totchym	2459	629·6								
Diet: Effect of:	SMP feed intake within diet		SPC feed intake within diet		SBM feed intake within diet		FM feed intake within diet		Diet	
	b	*P*	*b*	*P*	*b*	*P*	*b*	*P*	*P*	
Day 3 ('eaters')										
Pchym	−11·85	NS	−0·07	NS	−8·20	0·074	10·94	< 0·001	0·015§	
Jchym	1·98	NS	−0·48	NS	0·26	NS	0·15	NS	NS	
Pchymtot	−55·26	NS	−3·24	NS	−22·62	NS	32·62	0·002	0·018‖	
Jchymtot	2·49	NS	−3·97	0·075	9·15	0·026	2·54	NS	0·035¶	
totchym	−52·78	NS	−7·21	NS	−13·48	NS	35·16	0·002	0·22††	
	Pre-diet		Diet						Feed intake	
	LSM	SEM	SMP	SPC	SBM	FM	*P*	SEM	*b*‡	*P*
Day 6										
Pchym			432	532	442	269	NS	70·5	1·52	0·023
Jchym			154^{b}	95^{a}	104^{ab}	51^{a}	0·009	17·9	0·12	NS
Pchymtot			1230^{ab}	1913^{b}	1088^{a}	574^{a}	0·009	230·0	7·84	0·001
Jchymtot			810	850	846	508	NS	150·1	2·24	0·099
totchym			2040^{bc}	2763^{c}	1934^{b}	1082^{a}	0·003	251·4	10·08	< 0·001
Day 10										
Pchym			536	556	399	793	NS	93·9	1·75	0·064
Jchym			124	102	114	84	NS	13·5	−0·11	NS
Pchymtot			1803	2190	1285	2368	NS	343·3	8·25	0·021
Jchymtot			1195	868	1159	580	NS	172·5	0·48	NS
totchym			2999	3058	2443	2947	NS	330·1	8·73	0·013

LSM, least square mean; SMP, skimmed-milk powder; SPC, soya-bean-protein concentrate; SBM, soya-bean meal; FM, fish meal; Pchym, chymotrypsin activity/g freeze-dried pancreatic tissue; Jchym, chymotrypsin activity/g freeze-dried jejunal digesta; Pchymtot, total chymotrypsin activity in pancreas; Jchymtot, total chymotrypsin activity in jejunum; totchym total chymotrypsin activity in pancreas + jejunum; NS, not significant.

a, b, c Least square means within a row without a common superscript were significantly different ($P < 0·05$).

* 1 unit = amount of enzyme required to hydrolyse 1 μmol substrate (*N*-benzoyl-L-tyrosine ethyl ester)/min at 25° and pH 7·8.

† For details of diets and procedures, see Table 1 and pp. 353–355.

‡ Coefficient of regression.

§ FM > SPC.

‖ FM > SPC, FM > SBM.

¶ SMP > FM.

†† FM > SPC, FM > SBM.

At day 3, trypsin activity in the jejunum was affected by protein source (Table 5). The highest trypsin activity was found with piglets fed on SMP and the lowest activity was noticed in piglets fed on SBM and FM.

A significant effect of feed intake within diet on chymotrypsin activity at day 3 post-

Table 7. *Comparison of trypsin and chymotrypsin activities (units)* in the pancreas and jejunal digesta from newly-weaned piglets that consumed more than 100 g feed/d ('eaters') or less than 50 g feed/d ('non-eaters') in the 3 d immediately after weaning*†

(Least square means with their standard errors)

	'Eaters' (*n* 15)		'Non-eaters' (*n* 4)		
Day 3	LSM	SE	LSM	SE	*P*
Trypsin activity:					
Ptryp	1225	177	3029	342	< 0·001
Jtryp	268	51	150	98	NS
Ptryptot	2963	500	9239	969	< 0·001
Jtryptot	1547	284	370	549	0·074
tottryp	4510	660	9608	1277	0·003
Pt/c	2·85	0·31	1·08	0·61	0·019
Jt/c	4·02	0·36	0·84	0·70	0·001
Chymotrypsin activity:					
Pchym	467	70	2868	135	< 0·001
Jchym	63	10	150	19	< 0·001
Pchymtot	1138	278	8992	539	< 0·001
Jchymtot	369	52	199	100	NS
totchym	1507	274	9191	530	< 0·001

LSM, least square mean; Ptryp, trypsin activity/g freeze-dried pancreatic tissue; Jtryp, trypsin activity/g freeze-dried jejunal digesta; Ptryptot, total trypsin activity in pancreas; Jtryptot, total trypsin activity in jejunum; tottryp, total trypsin activity in pancreas+jejunum; Pchym, chymotrypsin activity/g freeze-dried pancreatic tissue; Jchym, chymotrypsin activity/g freeze-dried jejunal digesta; Pchymtot, total chymotrypsin activity in pancreas; Jchymtot, total chymotrypsin activity in jejunum; totchym, total chymotrypsin activity in pancreas+jejunum; Ptc/, Ptryp/Pchym; Jt/c, Jtryp/Jchym; NS, not significant.

* 1 unit = the amount of enzyme required to hydrolyse 1 μmol substrate/min at 25°.

† For details of diets and procedures, see Table 1 and pp. 353–355.

weaning was found (Table 6). Chymotrypsin activity in pancreatic tissue was positively related to feed intake only for piglets on the FM diet. At day 3, pancreatic chymotrypsin activity and total chymotrypsin activity (pancreas+jejunum) were higher for the FM-fed piglets than for the soya-bean-fed piglets. Jejunal chymotrypsin activity was higher for piglets fed on SMP than for piglets fed on FM.

At day 6, enzyme activities in pancreatic tissue and jejunal chyme were lowest for piglets fed on the FM diet compared with the other diets. Total enzyme activities (pancreas+jejunum) were highest for the SPC-fed piglets. Trypsin activity/g freeze-dried jejunal digesta was lower in piglets fed on FM than in piglets fed on SMP (Table 5). Chymotrypsin activity/g freeze-dried jejunal chyme was lower in piglets fed on FM and SPC than in piglets fed on SMP. Total enzyme activities in the pancreas were highest for SPC-fed piglets and lowest for FM-fed piglets (Tables 5 and 6). Total trypsin and chymotrypsin activities in pancreas+jejunum at day 6 were positively related to post-weaning feed intake. This was mainly due to the strong relationship between feed intake and pancreatic enzyme activities (total and per g pancreatic tissue).

At day 10 after weaning no effects of dietary protein source on either trypsin or chymotrypsin activities were found. In particular, the pancreatic trypsin and chymotrypsin activities seemed to increase sharply for the FM diet, resulting in equal pancreatic enzyme activities for all four diets.

DISCUSSION

Feed intake and growth

The low feed intake of piglets on the SMP diet may be related to the physical form of the feed. Feeds containing large amounts of skimmed-milk powder are difficult to pellet and will therefore result in hard pellets which are not readily accepted by young piglets (Jensen, 1966; Liptrap & Hogberg, 1991).

Piglets fed on the SMP diet consumed less feed during the first 3 d post-weaning than piglets fed on the FM diet (Table 2) and at the same time had higher gastric digesta weights (Table 3). This may indicate that emptying of gastric digesta was faster in piglets fed on FM, as was also found by Sève & Laplace (1975) with early-weaned piglets fed on solid diets containing milk and fish proteins. This could have resulted in a more regular feed intake for piglets fed on the FM diet.

The differences in feed intake between diets did not result in differences in average daily gain during the first 3 d post-weaning. Piglets fed on the diets based on animal proteins (SMP and FM) seemed to gain some live weight during the first 3 d post-weaning although the differences between diets were not significant. Average daily gain agreed with the results of Bark *et al.* (1986), who studied feed intake during the first week post-weaning in piglets weaned at 21 d of age.

Tissue weights

The relative weight of the empty stomach increased gradually after weaning, while the relative weight of the small-intestinal tissue decreased from weaning until 3 d after weaning. The same trends were found by Kelly *et al.* (1991*a*) with piglets weaned at 14 d of age. The initial post-weaning decrease in relative small-intestinal weight may be related to the decline in feed intake in these animals compared with intake during the suckling period. It could also indicate post-weaning morphological gut wall changes, because it was found that all piglets at day 6 had tongue- and leaf-shaped jejunal villi.

The (relative) pancreatic weight of SPC- and SMP-fed piglets increased from weaning until day 10. This is in accordance with the results of Kelly *et al.* (1991*a*) with piglets weaned at 14 d of age and fed on a diet containing skimmed-milk powder, fish meal and soya-bean meal. Piglets fed on the SBM diet had the lowest relative pancreatic weight at day 3 and piglets fed on the FM diet at day 6. The low pancreatic weight may be associated with the relatively (although not always significantly) low enzyme activities with the SBM- and FM-fed piglets on days 3 and 6 respectively.

Digesta pH

The increase in stomach pH after weaning is in accordance with the results of Wilson & Leibholz (1981*b*) and Efird *et al.* (1982). This result can be explained by post-weaning feed intake pattern and/or the buffering capacity of the post-weaning diets. A high pH in the stomach could lead to bacterial proliferation (Banwart, 1981) and to disturbance of normal pepsin (*EC* 3.4.23.1) function (Kidder & Manners, 1978).

At day 10, piglets fed on the soya-bean diets had lower pH in gastric contents than piglets fed on the animal protein sources. This could be explained by the buffering capacity of the diets, which was lower for the soya-bean diets (Table 1).

Gastric PP:CP ratio

The changes in gastric PP:CP ratio after weaning indicate that the degree of gastric protein breakdown is higher for solid diets than for sow's milk. This could be explained by the predominance of chymosin (*EC* 3.4.23.4) compared with pepsin (*EC* 3.4.23.1) activity in sucking piglets. Before weaning, coagulation of milk proteins (clot formation) is more important than protein hydrolysis (Cranwell & Moughan, 1989). Between days 3 and 10 an increase in gastric PP:CP ratio was found, indicating a decrease in gastric protein breakdown and/or an increase in gastric emptying rate of soluble protein.

At day 10 the gastric PP:CP ratio was lower for the FM-fed piglets than for the SPC-fed piglets, indicating a higher degree of gastric protein breakdown with the FM diet, since it was suggested earlier that gastric emptying rate was not decreased with the FM diet.

It is clear that the extent of gastric protein hydrolysis depends on age (or time after weaning) and dietary composition (protein source). This was also found by Leibholz (1986) who reported that piglets aged 28 d had higher stomach pH and less gastric protein breakdown than older piglets.

Gut wall morphology

The tongue- and leaf-shaped villi at day 6 are a common finding in newly-weaned piglets (Hampson, 1986; Deprez *et al.* 1987; Cera *et al.* 1988). Villus lengths were comparable with those reported by Deprez *et al.* (1987) of piglets at day 6 after weaning onto a dry diet, and by Hampson (1986) with piglets at day 5 after weaning, and by Miller *et al.* (1986) with piglets at 1 week after weaning. Crypt depths were slightly smaller than those reported by Hampson (1986), Miller *et al.* (1986), Deprez *et al.* (1987) and Kelly *et al.* (1991*a*).

The positive relationship between jejunal villus lengths and feed intake has been described previously by Kelly *et al.* (1991*b*). These authors used intragastric-tube feeding to establish different feeding levels in newly-weaned piglets. Villus lengths and crypt depths were positively related to feed intake (Kelly *et al.* 1991*b*) as also found in the present experiment. This relationship could indicate a stimulatory effect of feed intake on development of the gut wall.

It was proposed by Gall & Chung (1982) in rabbits and by Cera *et al.* (1988) in pigs that low feed intake during the early post-weaning period may be a contributing factor to reduced villus height. However, Kelly *et al.* (1991*b*) also found reductions in villus height in piglets fed through gastric intubation to maintain continuous nutrient supply.

Enzyme activities

The development of trypsin and chymotrypsin activities in pancreas and jejunum after weaning strongly depended on dietary protein source and post-weaning feed intake. At day 3, 'non-eaters' apparently stored large amounts of trypsin and chymotrypsin in their pancreatic tissue without substantial secretion into the gut. Intestinal substrate availability seems to be involved in the stimulation of pancreatic trypsin and chymotrypsin secretion as was suggested by DiMagno *et al.* (1973), Niederau *et al.* (1986) and Valette *et al.* (1992). This mechanism is stimulated through the digestive endproducts of intestinal protein digestion (Grendell & Rothman, 1981; Valette *et al.* 1992). DiMagno *et al.* (1973) found that essential amino acids infused into the duodenum or jejunum of humans stimulated pancreatic enzyme secretion. They postulated that the products of protein digestion after absorption inhibit pancreatic enzyme secretion through glucagon release. The study of Niederau *et al.* (1986) showed that arginine and lysine (the sites of tryptic cleavage) specifically caused the release of trypsinogen in a pancreatic tissue homogenate whereas phenylalanine and tryptophan (sites of chymotryptic cleavage) caused release of trypsinogen and chymotrypsinogen. Valette *et al.* (1992) found in experiments with pancreas-cannulated growing pigs fed on diets based on either casein or rape-seed that dietary protein source influences pancreatic enzyme secretion.

Skimmed-milk powder appeared to be the strongest stimulant of trypsin synthesis and secretion during the first 3 d post-weaning. At day 6, high pancreatic and jejunal trypsin activities were found with the SPC-fed piglets, while at day 10 the effect of dietary protein source on trypsin activities had disappeared. Feed intake was positively related to pancreatic trypsin activity and therefore might have affected trypsin synthesis.

The interaction between dietary protein source and feed intake during the first 3 d post-weaning with respect to pancreatic chymotrypsin activities is striking. Only piglets fed on FM (i.e. piglets consuming on average more than 100 g feed/d during the first 3 d post-weaning showed a positive relationship between feed intake and pancreatic chymotrypsin activity.

Piglets fed on the SMP diet had low feed intakes but higher gastric digesta weights compared with piglets fed on the FM diet. This suggests a higher rate of gastric emptying when FM was fed. A faster gastric emptying combined with a higher gastric pH leads to a more regular supply of more alkaline digesta to the duodenum. From this it can be expected that the pancreas is less challenged to secrete bicarbonate into the gut lumen. This hypothesis is supported by the finding that pancreatic tissue weight was also lower for piglets fed on the FM diet. The low enzyme activities in the jejunum of piglets fed on FM could reflect a lower need for pancreatic enzymes when gastric emptying occurs more gradually (i.e. when the FM diet is fed). It is evident that piglets fed on the FM diet had the highest feed intake during the first 3 d post-weaning. This could imply a more regular development of feed intake of newly-weaned piglets fed on the FM diet. This regular increase in feed intake after weaning may also imply a better development of the enzyme system, reflected by the positive relationship between feed intake and enzyme activities associated with the FM diet. At day 6 the lowest chymotrypsin activities were found with the FM diet and the highest with the SMP and SPC diets. By day 10 the differences between diets had disappeared, while feed intake was still related to total pancreatic chymotrypsin activity. The trypsin:chymotrypsin ratio in pancreas and jejunum was lower for 'non-eaters' than for 'eaters'. This is in accordance with the findings of Corring *et al.* (1978), Efird *et al.* (1982), Owsley *et al.* (1986) and Lindemann *et al.* (1986) who state that chymotrypsin is the predominant pancreatic protease during the suckling period, while trypsin increases specifically after weaning. Our results indicate that the shift from chymotrypsin to trypsin may be related to post-weaning solid feed intake rather than to weaning itself.

CONCLUSIONS

During the first 3 d post-weaning, feed intake was affected by dietary protein source. Dietary FM had a stimulatory effect on the development of post-weaning feed intake, probably related to modifications of gastric emptying patterns which may in turn be related to intestinal protein digestion. SPC diet was associated with high pancreatic tissue weight. Dietary protein source affected the development of trypsin and chymotrypsin activities in pancreatic tissue and jejunal digesta after weaning. Dietary FM generally resulted in low enzyme activities, while SMP and SPC were associated with high enzyme activities. Feed intake during the early post-weaning period clearly stimulated the development of the pancreas and the gut wall.

Dietary buffering capacity, gastric protein hydrolysis and gastric emptying seem to be important factors in the digestion of different protein sources by newly-weaned piglets. From the results presented herein it can be derived that feed intake as well as dietary composition are important factors for the development of the digestive organs of newly-weaned piglets.

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