Artificial rearing of pigs

9. Effect of replacement of dried skim-milk by fish-protein concentrate on performance and digestion of protein

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1. Half (diet M) or total replacement (diet N) of dried skim-milk by fish-protein concentrate and dried whey was compared with a diet containing dried skim-milk (710 g/kg) as the only source of protein (diet A). All three diets were prepared as spray-dried powders, and contained 20 g starch and 270 g soya-bean oil/kg. The diets were calculated to be isonitrogenous, but upon analysis had crude protein (nitrogen × 6·25) contents (g/kg) of 230, 270 and 291 in diets A, M and N respectively.

2. The diets were reconstituted as liquids containing 200 g dry matter (DM)/l and given to pigs weaned at 2 d of age during a 26 d experiment. The diets were fed at hourly intervals to a scale based on live weight. Protein digestion was studied in pigs, 6 d of age, and killed 1 h after a feed.

3. Live-weight gain was slightly improved when half the dried skim-milk was replaced, but the feed: gain ratio (g DM consumed/g live weight gain) was unaltered. Total replacement of dried skim-milk, markedly reduced performance and increased the incidence of scouring and mortality.

4. Increasing the proportion of fish protein reduced the apparent digestibility (AD) and the retention of N (g/d per kg live weight). N retention, but not AD, decreased with age.

5. Total replacement of dried skim-milk decreased the amount of digesta in the stomach, its pH value, and the proportion of DM and total N in the digesta, but non-protein N as a proportion of total N was increased.

6. The total pepsin activity in the stomach digesta was decreased by the substitution with fish protein, but there was no change in the activity of the stomach tissue. Trypsin and chymotrypsin activities in the pancreas were not affected by the source of dietary protein, but in the digesta of the small intestine the activity of these two enzymes was markedly reduced when the diet contained fish protein.

7. Total replacement of dried skim-milk with fish-protein concentrate and dried whey may increase the rate of flow of digesta through the small intestine, which together with the reductions in the apparent digestibility of N and amounts of trypsin and chymotrypsin in the digesta could reduce the efficiency of protein digestion and adversely affect performance.

In artificial rearing, pigs are weaned at birth, or within a few days of birth. Before this technique could be applied under commercial conditions, solutions are required to three major problems: satisfactory reproductive performance in sows after weaning, control of scouring in artificially-reared pigs, and the development of a milk-substitute which satisfies the nutritional requirements of the baby pig. These problems have been discussed in a recent review (Newport, 1977).

Although excellent performance can be obtained by feeding pigs weaned at 2 d of age on diets containing dried skim-milk as the only source of protein (Braude & Newport, 1973), under commercial conditions, the large-scale use of skim-milk for this purpose would be uneconomical, except perhaps for the first few days after weaning. Therefore, alternative sources of protein for artificially-reared pigs must be evaluated.

In the present experiment, a fish-protein concentrate has been studied. This was produced by controlled proteolysis of fish offal, followed by centrifugation to remove insoluble material. The soluble fraction was then defatted, concentrated and spray-dried. Seve *et al.* (1975) reported that only one-third of the dietary protein, supplied by skim-milk could be replaced by fish-protein concentrate for pigs weaned at 12 d of age. Pond, Snyder *et al.*

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Table 1. *Composition of the spray-dried diets*

	Diet		
	A	M	N
Ingredients (g/kg)			
Dried skim-milk	710	360	—
Dried whey	—	230	470
Fish-protein concentrate†	—	120	240
Soya-bean oil	270	270	270
Starch	20	20	20
Chemical analysis (g/kg)			
Crude protein (nitrogen × 6·25)	230	270	291
Total lipid	326	304	288
Calcium	9·6	7·1	4·8
Phosphorus	7·8	7·1	6·0
Amino acids:			
Lysine	17·1	19·5	19·2
Histidine	6·1	6·4	5·6
Arginine	7·3	13·9	15·2
Tryptophan‡	2·9	3·0	2·9
Threonine	10·5	11·2	12·2
Methionine + cystine	6·9	7·7	8·4
Valine	14·2	12·5	13·4
Isoleucine	12·6	12·7	12·0
Leucine	22·1	22·2	20·2
Tyrosine	12·0	10·6	8·4
Phenylalanine	11·6	11·3	10·0
Aspartic acid	18·6	21·3	24·3
Serine	13·2	14·4	13·5
Glutamic acid	50·3	47·2	41·1
Proline	22·3	19·5	15·8
Glycine	5·0	13·4	22·0
Alanine	7·7	12·5	16·8

† Obtained from Cooperative de Traitment des Produits de la Peche, Boulogne, France.
‡ Calculated values.

(1971) found that fish-protein concentrate was equal to casein as a source of protein for pigs weaned at 2 d of age, but in their experiment food intakes, and consequently growth, were very low (approximately 60 g/d from 2–23 d of age). In the experiment now reported, the effect of half or total replacement of dried skim-milk by a mixture of fish-protein concentrate and dried whey has been studied in pigs weaned at 2 d of age. The digestion of the two types of protein has also been studied in an attempt to elucidate the reasons for the adverse effect on performance which was found when there was total replacement of dried skim-milk.

EXPERIMENTAL

Diets and feeding scale

Three spray-dried powders, described in Table 1, were prepared from mixtures of skim-milk and soya-bean oil alone, or in combination with fish-protein concentrate and dried whey. Starch (20 g/kg) was added to all three diets. (Under the current policy of the European Economic Community the cost of dried skim-milk without a 'denaturing' agent is considerably greater than 'denatured' skim-milk. Starch is an approved 'denaturing' agent). Spray-drying was carried out using a mild-heat process to prevent the denaturation of the whey proteins in the skim-milk. Liquid diets containing 200 g dry matter (DM)/l, and supplemented with retinol, cholecalciferol, α-tocopherol and phylloquinone were prepared as previously described (Braude & Newport, 1973). Diets containing fish protein (diets M and N) were also supplemented with solutions containing 201 g $NaH_2PO_4.2H_2O$/l and 548 g $CaCl_2.6H_2O$/l to increase the phosphorus and calcium contents of these two diets (on a DM basis) to that of the milk-protein diet (diet A).

The pigs were fed at hourly intervals on the scale described by Braude & Newport (1973).

Experimental design

Litter-mate, 2-d-old pigs were allocated to the three treatments (diets) on the basis of live weight and sex. The pigs on different treatments were kept in separate rooms, each containing four replicates. This procedure was repeated six times so that there were twenty-four pigs/treatment. The pigs were on experiment until 28 d of age. During the experiment, nine litter-mate blocks were discarded after two, or all three pigs, died following a period of scouring. Thus results for fifteen pigs/treatment were available for analysis. In these results there were six missing values, again due to the death of the pigs after scouring. The results were analysed after missing values had been estimated.

A further twelve pigs/treatment were weaned at 2 d of age, and killed at 6 d of age in a study of the digestion of the dietary proteins.

Nitrogen retention and procedure at slaughter

N retention was estimated from a collection period of 4 d duration, as previously described (Braude *et al.* 1976). Collection periods were not attempted when pigs were scouring, and spillage of diet into the urine further reduced the number.

In the study of digestion, the pigs were killed 1 h after a feed by an intracardiac injection of sodium pentobarbitone. The stomach and pancreas were removed, the digesta emptied from the stomach and its pH determined with a glass electrode. The small intestine was removed and the digesta washed from it using a solution of sodium chloride (9 g/l) at room temperature (approximately 20°). The stomach wall, pancreas and digesta were stored at −20° until analysed.

Analytical methods

The determinations of DM, total and non-protein-N and total lipid have been described by Braude *et al.* (1970) and Braude & Newport (1973). After ashing samples of diet, Ca was determined by atomic absorption spectroscopy, and P by colorimetric estimation of the phosphovanadomolybdate complex (Cavell, 1955). The amino acid composition of the diets was determined using methods described in Braude *et al.* (1977). The pepsin content of the stomach tissue and digesta was assayed by the method of Anson (1938). One unit of pepsin activity was defined as an increase in extinction at 280 mμ of 0·001/min at 37°. Trypsin and chymotrypsin were estimated in the pancreas and digesta in the small intestine as described by Hummel (1959). The pancreatic enzymes were assayed after activation with enterokinase (*EC* 3.4.21.9) (Miles Laboratories Ltd, Stoke Poges). The activity of both trypsin and chymotrypsin were calculated by comparison with the purified enzymes (Koch-Light Laboratories, Colnbrook, Bucks.).

RESULTS

Performance

The performance of the pigs is shown in Table 2. Total replacement of dried skim-milk by fish-protein concentrate and dried whey severely reduced performance ($P < 0·001$). Replacement of half the dried skim-milk improved growth rate ($P < 0·01$), and decreased the feed: gain ratio, although this decrease was not statistically significant ($P > 0·05$).

Table 2. *Effect of replacing dried skim-milk by fish-protein concentrate and dried whey on performance of pigs from 2–28 d of age*

(Mean values for fifteen pigs/treatment)

Diet† Replacement of dried skim-milk (%) ...	A 0	M 50	N 100	SE of a mean (22 df)	Statistical significance of differences between diets
Live-wt gain (g/d)	216	249	130	7·3	M > A**, A, M > N***
Feed:gain (g dry matter consumed/g live-wt gain)	0·99	0·95	1·28	0·021	N > A, M***

Two pigs given diet A, and four pigs given diet N died. Missing values were calculated.
** $P < 0·01$, *** $P < 0·001$.
† For details of diet composition, see Table 1.

Table 3. *Effect of replacing dried skim-milk by fish-protein concentrate and dried whey on the apparent digestibility (AD) of nitrogen in pigs between 2 and 28 d of age*

(Results are mean values with their standard errors)

Diet† ... Replacement of dried skim-milk (%) ...	A 0	M 50	N 100	Statistical significance of differences between diets
No. of pigs	4	12	13	
AD	0·998 ±0·001	0·988 ±0·002	0·975 ±0·004	A > M > N*

* $P < 0·05$.
† For details of diet composition, see Table 1.

Apparent digestibility (AD) and retention of N

AD was estimated between 7 and 26 d of age (mean age 20 d). The results are given in Table 3. AD was reduced by fish protein and the effect, although small, was statistically significant ($P < 0·05$). There was no effect of age on AD.

N retention (g/d per kg live weight) decreased with age (Fig. 1), and was lower ($P < 0·01$) in pigs given the all-fish-protein diet (diet N) compared with the milk-protein diet (diet A).

Digestion of protein in the stomach

The amount of digesta in the stomach of pigs slaughtered at 1 h after a feed was reduced when fish protein replaced all the dried skim-milk, but replacing half the dried skim-milk had no effect (Table 4). Increasing the amount of fish protein in the diet, progressively reduced the DM content, and total replacement of dried skim-milk resulted in digesta with no discernible clot. A small decrease in pH was also observed as dried skim-milk was replaced. The proportion of total N in the digesta in the form of non-protein-N was considerably increased by the fish-protein diets.

The concentrations of pepsin in the stomach tissue and digesta were not affected by the source of protein, except for a decrease in the pepsin concentration in the digesta from pigs fed on the diet with half the dried skim-milk replaced (Table 5).

Digestion of protein in the small intestine

There was some reduction in the weight of the pancreas/kg live weight in pigs given the fish-protein diets, although the concentration of trypsin and chymotrypsin in the pancreas was not affected by the source of dietary protein (Table 5). However, the amounts of trypsin

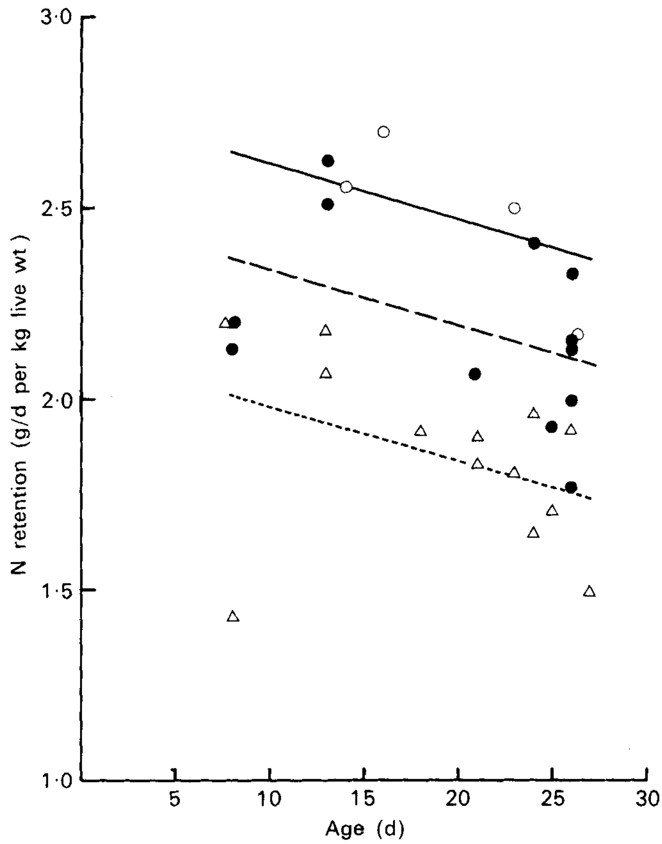

Fig. 1. Effect of age (d) and source of dietary protein on nitrogen retention (g/d per kg live weight) in pigs given diets in which fish-protein concentrate (diets M and N) replaced dried skim-milk (diet A) (for details of diets, see Table 1 and p. 104. ○—○, Diet A; ●---●, Diet M; △- - -△, Diet N.

Table 4. *Effect of replacing dried skim-milk by fish-protein concentrate and dried whey on the amount and composition of digesta in the stomach of* 6-*d-old pigs* 1 *h after feeding*

Diet†	No. of pigs	A	M	N	SE of a mean	df	Statistical significance of differences between diets
Replacement of dried skim-milk (%) ...		0	50	100			
Total wt (g)	10	97·1	94·9	31·4	7·40	20	A, M > N***
Dry matter (g/kg)	10	139·0	96·8	70·8	6·14	20	A > M, N***; M > N**
pH	10	4·13	3·90	3·66	0·188	20	NS
Total N (mg/g wet wt)	8	14·0	8·5	6·1	0·62	14	A > M, N***; M > N*
Non-protein N (mg/g wet wt)	8	0·82	1·38	1·87	0·055	14	N > M > A***
Non-protein N (% total N)	8	5·9	16·2	30·7	—	—	—

NS, not significant ($P > 0·05$).
* $P < 0·05$, ** $P < 0·01$, *** $P < 0·001$.
† For details of diet composition, see Table 1.

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Table 5. *Effect of replacing dried skim-milk by fish-protein concentrate and dried whey on the concentration of pepsin in stomach digesta and tissue, and the concentration in the pancreas and amount in the digesta in the small intestine of trypsin and chymotrypsin in six-day-old pigs*

(Mean values for twelve pigs/treatment)

Diet† ...	A	M	N	—		—
Replacement of dried skim-milk (%) ...	0	50	100	SE of a mean	df	Statistical significance of differences between diets
Pepsin:						
In digesta (units/g)	22·5	12·0	21·6	3·54	20‡	A, N > M*
Total digesta (units)	2045	971	991	388	18	NS
Stomach tissue (units/g)	178	169	162	30·2	20‡	NS
Pancreas (g/kg live wt)	1·75	1·29	1·34	0·070	22	A > M, N***
Trypsin:						
In pancreas (mg/g)	0·87	0·79	0·69	0·077	22	NS
Total in digesta (mg)	10·4	3·1	4·7	1·77	22	A > M**, > N*
Chymotrypsin:						
In pancreas (mg/g)	2·67	2·62	2·13	0·353	22	NS
Total in digesta (mg)	47·3	25·8	27·0	6·35	22	A > M, N*

NS, not significant ($P > 0·05$).
* $P < 0·05$, ** $P < 0·01$, ***$P < 0·001$.
† For details of diet composition, see Table 1.
‡ Missing values were given for two pigs given diet N.

and chymotrypsin in the digesta from the small intestine were considerably reduced when the diet contained fish protein. Trypsin was not detected in digesta from six pigs given diet M (500g fish protein/kg protein) and five pigs given diet N (all fish protein).

DISCUSSION

All three diets were intended to be isonitrogenous, based on calculated values for the ingredients. However, analysis of the diets showed that crude protein (N × 6·25; CP) level increased as fish protein was substituted for dried skim-milk. Thus, the CP content of the diet M (500 g fish protein/kg protein) was greater (270 g CP/kg DM) than that of diet A (all milk) (240 g CP/kg DM), and this difference might account for the higher growth rate found when pigs received diet M, as in a previous experiment (Newport, 1978), a similar difference in growth rates was found in response to dietary CP level. Although performance was not adversely affected by replacing half the milk protein in the diet by fish-protein concentrate, the results clearly show that when dried skim-milk was totally replaced, performance was very poor and was associated with a higher incidence of scouring and mortality.

The reduction in N retention (g/d per kg live weight) with age (Fig. 1) has also been found when diets of liquid or spray-dried whole cow's milk were given (Braude *et al.* 1970). The individual linear regression analysis for each diet was not statistically significant ($P > 0·05$), probably due to the limited results available, particularly for diet A (all milk). When the pooled results were analysed, a statistically significant ($P < 0·01$) reduction in N retention was found when fish (diet N) replaced milk protein (diet A). The regression lines shown in Fig. 1 were fitted using a common slope, as the slopes of the individual regressions did not differ significantly ($P > 0·05$). The effect of fish protein on N retention cannot be accounted for by the small decrease in the apparent digestibility ratio (Table 3), and was mainly due to a greater loss of N in the urine. The true difference in the AD of N between the diets is probably underestimated as observations were only made on healthy pigs, whereas scouring was more severe in pigs given diet N (all fish protein).

The reduction in AD may be due to an increase in the rate of flow of digesta through the small intestine, resulting in less efficient absorption. Although this experiment did not provide any direct evidence for this, the decreased amount of digesta in the stomach of pigs fed on the diet containing no dried skim milk (Table 4) indicated that rate of stomach emptying, and possibly flow through the small intestine may have increased. The all-fish-protein diet did not coagulate in the stomach, a factor which may increase the rate of stomach emptying. An increase in the rate of flow of digesta might contribute to the greater severity of the scouring observed in pigs given diet N (all fish protein).

There was a much greater proportion of non-protein-N relative to total N in the digesta from the stomach of pigs given fish protein compared with dried skim-milk. This difference cannot be due only to the greater content of non-protein-N in the diet N (8·5 % of the total N) compared with the diet A (all milk) (4·2 % of the total N), and suggests fish protein may be more readily hydrolysed, or partially hydrolysed, by pepsin than is milk protein.

The concentration of pepsin in the stomach digesta was reduced by approximately 50 % when half the dried skim-milk was replaced by fish protein (Table 5). Similar results have also been reported by Seve & Laplace (1975). The concentration of pepsin in the diet N was however higher and similar to that in the diet A. The total pepsin content of the digesta was reduced when fish protein replaced dried skim-milk, suggesting that the secretory response to the fish protein diets (diets M and N) may be lower than for milk protein which is in accordance with the results indicating a greater ease of hydrolysis as indicated by the proportion of non-protein-N relative to total N in the digesta.

The results also suggest that pancreatic secretion into the small intestine may be reduced by fish protein. Although the concentrations of trypsin and chymotrypsin in the pancreas were not affected, there was a reduction in the weight of the pancreas in pigs given diets M and N. Thus, the total amounts of these enzymes were less in the pancreas of pigs fed on fish protein and may indicate that the rate of pancreatic secretion into the small intestine may be impaired which would also explain the marked reduction in the amounts of trypsin and chymotrypsin present in the digesta from the small intestine. Pond *et al.* (1971) reported that neither the weight of the pancreas, nor the concentrations of trypsin and chymotrypsin in the pancreas were affected by replacing casein with fish-protein concentrate. The similar effect of the two protein sources may have been due to the slow growth rates, arising from the low level of feeding in that experiment (Pond *et al.* 1971). A reduction in pancreatic secretion in combination with an increased rate of flow through the small intestine could result in the incomplete hydrolysis, and absorption, of the partially-digested protein. If fish protein was less amenable to intestinal proteolysis than milk protein, a reduction in pancreatic secretion would explain the poor performance of pigs given the all-fish-protein diet, compared with those pigs given an all-milk diet.

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