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Requirement of young pigs for apparent ileal digestible tryptophan

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Abstract

Five growth trials involving a total number of 420 young pigs were performed to study the tryptophan requirement during the live weight range of 20 to 40 kg. Five levels of L-tryptophan addition (0.0, 0.15, 0.30, 0.45 and 0.60 g kg^{-1}) were evaluated using a basal control diet, containing 162 g kg^{-1} crude protein (CP) and 1.65 g kg^{-1} tryptophan. The control diet was supplemented with lysine, methionine + cystine, threonine, isoleucine, valine and histidine to assure that only tryptophan was limiting pig performance.

The experimental diets were fed ad libitum as pellets. The requirement for total tryptophan was found to be approximately 2.10 g kg^{-1} for optimal weight gain and maximum efficiency of feed utilization, in a diet containing 9.5 MJ kg^{-1} net energy. This value corresponds with approximately 1.77 g kg^{-1} apparent ileal digestible tryptophan.

Keywords: pigs, tryptophan, requirement, digestibility

Introduction

In several West European countries pollution of the environment with nitrogen originating from animal manure is becoming a major problem. Consequently the utilization of dietary protein is currently undergoing critical assessment. One of the most important factors affecting the protein utilization is the balance of amino acids in the feed protein. The closer the amino acid composition of the diet matches the requirement for maintenance and production, the less protein the animal effectively needs. In pig diets the most critical amino acids are lysine, methionine, and threonine. Therefore, supplementation of diets for growing pigs with these amino acids provides a mean for increasing the efficiency of the utilization of dietary protein (Jongbloed & Lenis, 1992; Schutte & Tamminga, 1992). After these amino acids, tryptophan may often become limiting, in particular when the diet contains large

proportions of maize and meat meal. Therefore, the pig's requirement for ileal digestible tryptophan should be known accurately.

There is a wide variation in recommended requirements for tryptophan in young pigs, due to differences in diet composition, weight gain, feed intake and in capacity for protein deposition among the pigs used in the studies. Moreover, most of the reported requirement figures for tryptophan are based upon total rather than on (ileal) digestible basis. In addition, some of the studies reported are difficult to interpret either because of the unreliability of the analytical method used for determination the tryptophan level in feeds and feedstuffs, or the failure to consider the effects of the levels of other amino acids in the diets on performance to tryptophan additions (Sato *et al.*, 1987).

The ARC (Anonymous, 1981) recommends a dietary level of 1.8 g kg^{-1} of tryptophan for pigs during the age period of 3 to 8 weeks and 1.6 g kg^{-1} for pigs in the weight range of 15-50 kg (8.8 MJ net energy kg^{-1} i.e. 12.6 MJ ME kg^{-1}). The NRC (Anonymous, 1988) recommends a dietary tryptophan level of 1.4 g kg^{-1} and 1.2 g kg^{-1} for pigs in the weight range of 10 to 20 kg and 20 to 50 kg, respectively, when corn-soyabean meal diets (9.3 MJ kg^{-1} net energy i.e. 13.3 MJ ME kg^{-1}) are fed. The values for tryptophan recommended by ARC (Anonymous, 1981) and NRC (Anonymous, 1988) correspond with approximately 15 % of the dietary lysine content. This value is similar to that reported by Southern (1991) on an ileal digestible basis. Henry *et al.* (1986) reported maximum growth performance of pigs at 1.58 g kg^{-1} dietary tryptophan for the live weight period of 15 to 40 kg. Lewis *et al.* (1977) showed a significant interaction between the dietary lysine level and the requirement for tryptophan in piglets of 3 weeks of age. They found at lysine contents of 5.7, 7.7 and 9.7 g kg^{-1}, tryptophan requirements of 1.6, 1.9 and 2.3 kg^{-1}, respectively. Borg *et al.* (1987) found a requirement of 1.6 g kg^{-1} tryptophan in pigs for the live weight period of 6 to 22 kg using lysine contents of 10 and 11 g kg^{-1} in the diet. Russell *et al.* (1983) determined a tryptophan requirement of 1.7 g kg^{-1} for pigs of 18 to 35 kg of body weight with a dietary lysine content of 8.1 g kg^{-1}. Bertram & Berende (1983) established the highest performance of pigs in the weight range of 12 to 40 kg with 2.1 g tryptophan g kg^{-1} diet. The experimental diet contained 175 CP g kg^{-1} and 11.7 g kg^{-1} of lysine. A tryptophan requirement of 2.3 g kg^{-1} for pigs of 10 to 35 kg of body weight was determined by Schutte *et al.* (1988) with a diet containing 10.3 MJ kg^{-1} net energy (i.e. 14.7 MJ ME kg^{-1}) and 11.5 kg^{-1} lysine. Their estimated requirement figure for tryptophan was relative to the content of lysine, about 20%. This value corresponds well with those reported by Fuller *et al.* (1989) being 19% of the requirement level of lysine. A much higher estimate of tryptophan requirement was reported by Fremaut & de Schrijver (1990), who stated on the basis of daily gain, that total tryptophan requirement of growing pigs (20 to 50 kg) is 23% of total lysine requirement.

The study reported in this paper was carried out to determine the requirement for ileal digestible tryptophan of young pigs. 'Requirement' was defined as the marginal value above which additional supplementation of tryptophan did not further improve performance of pigs.

Materials and methods

The study involved a digestibility trial and five growth trials. The trials were performed at two institutes. Two of the growth trials and the ileal digestibility trial were carried out at the TNO-Institute of Animal Nutrition and Physiology (ILOB), Wageningen, The Netherlands, and the other three growth trials at the Institute for Animal Science and Health (ID-DLO), Lelystad, The Netherlands.

Diets

The composition of the basal diet, is presented in Table 1. The basal diet was analysed to contain 162 g kg^{-1} CP and 1.65 g kg^{-1} tryptophan. Five levels of addition of L-tryptophan (0.0, 0.15, 0.30, 0.45 and 0.60 g kg^{-1}) were tested, resulting in dietary tryptophan levels of 1.65, 1.80, 1.95, 2.10 and 2.25 g kg^{-1}, respectively. The basal diet was supplemented with lysine, methionine + cystine, threonine, isoleucine, valine and histidine in order to be sure that only tryptophan was limiting. Before inclusion in the diets, all feed ingredients, except tapioca, were analysed for contents of CP, Ca, P and amino acids. Based on these figures, the basal diet was formulated. The basal diet for all experiments was prepared as one batch. The basal diet was analysed for the contents of CP, Ca, P and amino acids (Table 2).

Table 1. Composition of the basal diet (g kg^{-1}).

Ingredients	
Tapioca	251.0
Barley	300.0
Corn	157.6
Soya bean meal	120.0
Corn gluten meal	35.0
Alfalfa meal	20.0
Skim milk powder	40.0
Cane molasses	20.0
Soya oil	19.0
Limestone	4.0
Dicalciumphosphate	17.5
Salt	3.0
Vitamin/mineral premix[1]	2.0
L-lysine HCL	5.1
DL-methionine	1.8
L-threonine	1.7
L-isoleucine	1.1
L-histidine	0.5
L-valine	0.7

[1] The vitamin/mineral premix supplied per 1 kg feed: 9000 IU vitamin A, 1800 IU vitamin D3, 40 mg DL-α-tocopheryl acetate, 5 mg riboflavin, 30 mg niacin, 12 mg d-pantothenic acid, 250 mg cholinechloride, 40 µg cobalamin, 3 mg menadione, 50 mg ascorbic acid, 0.3 mg folic acid, 160 mg Cu, 80 mg Fe, 73 mg Zn, 44 mg Mn, 0.5 mg Co, 0.06 mg Se, 0.4 mg I and 40 mg Tylosin.

Table 2. Chemical composition of the basal diet (in g kg^{-1}).

Crude Protein (CP)[1]	162
Dry matter (DM)[1]	887
Digestible energy (MJ kg^{-1})[2]	14.0
Metabolizable energy (MJ kg^{-1})[2]	13.6
Net energy (MJ kg^{-1})[2]	9.50
Ca[1]	8.00
P[1]	6.7
P[2] (available)	3.7
Amino acids[1]	
Isoleucine	7.7
Leucine	13.7
Lysine	10.9
Methionine	4.3
Cystine	2.6
Methionine + cystine	6.9
Phenylalanine	6.9
Tyrosine	5.3
Threonine	7.2
Tryptophan	1.65
Valine	8.5
Arginine	8.4
Histidine	4.0

[1] Analysed values
[2] Values calculated from data provided by the Dutch Bureau of Livestock Feeding (1994).

Experimental procedures

Ileal digestibility trial

The apparent ileal digestibility of protein and amino acids of the basal diet was determined using five castrated male pigs (GY × NL). Two pigs were provided with an ileo-caecal re-entrant cannula (20 mm internal diameter, silicone rubber), according to the technique developed by Easter & Tanksley (1973). The other three pigs were provided with a post valvular T-caecum (PVTC) cannula (Van Leeuwen *et al.*, 1991). The mean live weight of the pigs was approximately 50 kg. The pigs were individually housed in metabolism cages in an environmentally controlled unit with continuous light. Air temperature varied between 18 and 21 °C. Water was administered with the meal at a ratio of 2:1. The amounts of feed given were based on live weight of the animals and energy content of the diet. The feeding level was 2.5 times the energy requirement for maintenance corresponding with 1.05 MJ metabolizable energy/kg metabolic body weight. After a preliminary period of 3 days in which the animals received the same amount of the diet as in the collection period, the ileal digesta were collected quantitatively for 3 × 24 h on alternate days. Digesta were collected hourly, weighed and frozen (–20 °C).After the collection period a sample of the pooled digesta was freeze dried, ground and analysed for contents of DM, OM, CP, crude fat, CF and amino acids.

Growth experiments

The experimental treatments (Table 3) were similar in each of the five growth experiments.

The animals (GY × NL) were group-housed in an artificially heated, ventilated and lighted room. The temperature in the pig unit varied between 20 and 23 °C. After an acclimatisation period (14 days for the two ILOB trials, 7 days for the three IVVO trials), the animals were allotted to the five treatments, based on live weight, origin, sex and weight gain after weaning. In the ILOB trials, each experimental diet was fed to four replicate pens of 8 group-housed pigs each; two pens with barrows and two with gilts. In the IVVO trials, each diet was fed to one pen with 7 group-housed pigs (ratio barrows to gilts 3:4 or 4:3). The experimental diets were fed ad libitum as pellets for a period of four weeks. Drinking water was freely available. For all experiments, the mean weight of the animals at the start of the experimental periods was approximately 19 kg. At the end of the experimental period (after four weeks) the animals were weighed individually. Feed intake and feed conversion efficiency, calculated as kg feed/kg weight gain, were determined per replicate.

Chemical analysis

N content of the diets and faeces were determined according to the Kjeldahl method. CP content was calculated as N x 6.25. Amino acids with the exception of methionine, cystine and tryptophan, were determined by ion-exchange chromatography (Beck *et al.*, 1978) after hydrolysis of the samples with 6 M HCl for 24 h, and using correction factors for isoleucine and valine of Slump (1969). Methionine and cystine were determined as methionine sulphone and cysteic acid, respectively, after oxidation with performic acid according to Moore (1963). The oxidized samples were hydrolysed in the same way as the unoxidized samples. The amino acid composition of the hydrolysates were determined with an automatic amino acid analyzer (Biotronic LC 6001). Tryptophan was determined after hydrolysis with 2.7 N $Ba(OH)_2$ (8 h at 130 °C) according to Slump & Schreuder (1969).

Table 3. Experimental treatments in the growth trials.

Group	Added L-tryptophan (g kg^{-1})	Content of tryptophan (g kg^{-1})	
		total[1]	ileal digestible[2]
1	0	1.65	1.32
2	0.15	1.80	1.47
3	0.30	1.95	1.62
4	0.45	2.10	1.77
5	0.60	2.25	1.92

[1] Analysed contents

[2] The ileal digestibility is calculated using the digestibility coefficient of tryptophan measured in the digestibility trial. In this calculation the digestibility of the added L-tryptophan was assumed to be 100%.

Statistical analysis

The results obtained by ILOB and IVVO were analysed separately. The data on weight gain and feed conversion efficiency were analysed according to a randomized-block design with experiment, sex (only ILOB) and treatment as factors. Statistical significance of differences between treatments were determined with the Least Significant Difference (LSD) test (Snedecor & Cochran, 1980).

Results and discussion

The body weight of the pigs used in the digestibility trial was higher than in the growth trials (50 to 55 kg versus 20 to 40 kg). Based on the results of Schutte *et al.* (1992), however, it can be assumed that the ileal digestibility values of nutrients obtained in pigs with a body weight of 50 kg are transferable to pigs in the weight range of 20 to 40 kg. In the study of Schutte *et al.* (1992) no indications were found that ileal digestibility of nutrients is affected by age within the live weight period of 25 to 55 kg.

Apparent ileal digestibility values for DM, OM, CP, crude fat, crude fibre and amino acids are shown in Table 4. The ileal digestibility of cystine and tryptophan

Table 4. Ileal digestibility coefficients of dry matter, organic matter, crude protein, crude fat, crude fibre and amino acids in the basal diet (mean ± SD).

	Ileal digestiblity (%)
Dry matter	77.6 ± 2.9
Organic matter	80.5 ± 2.5
Crude protein	82.3 ± 2.9
Crude fat	88.8 ± 1.9
Crude fibre	25.3 ± 14.8
Isoleucine	87.7 ± 1.8
Leucine	89.9 ± 1.2
Lysine	88.8 ± 2.2
Methionine	93.1 ± 1.2
Cystine	74.0 ± 4.2
Phenylalanine	87.8 ± 1.8
Tyrosine	87.8 ± 1.9
Threonine	83.5 ± 2.6
Tryptophan	79.9 ± 3.2
Valine	85.3 ± 2.1
Arginine	89.1 ± 1.5
Histidine	85.3 ± 1.8
Alanine	84.1 ± 2.6
Aspartic acid	83.6 ± 2.3
Glutamic acid	88.4 ± 1.9
Glycine	75.6 ± 3.7
Proline	86.6 ± 2.3
Serine	84.7 ± 1.9

Table 5. Mean combined results of the two ILOB growth experiments for daily weight gain, feed conversion and daily feed intake.

Group	Tryptophan content (g kg^{-1})		Weight gain (g d^{-1})[1]	Feed conversion (kg feed kg^{-1} gain)[1]	Feed intake (g d^{-1})
	total[2]	ileal[3] digestible			
1	1.65	1.32	683^{a}	1.981^{b}	1,353
2	1.80	1.47	703^{ab}	1.928^{ab}	1,355
3	1.95	1.62	716^{ab}	1.920^{ab}	1,375
4	2.10	1.77	741^{b}	1.909^{a}	1,415
5	2.25	1.92	727^{ab}	1.909^{a}	1,388
LSD ($P < 0.05$)			47.0	0.067	

[1] Values within a column not having a common superscript differ significantly ($P < 0.05$).
[2] Analysed contents.
[3] Calculated using the digestibility coefficient for tryptophan determined in the digestibility trial.

was found to be slightly lower than the digestibility of most of the other amino acids and crude protein. These findings are in agreement with those reported by Sauer & Ozimek (1986) and Van Leeuwen *et al.* (1987).

The results of the growth trials performed at ILOB are summarized in Table 5. In both trials no significant ($P>0.05$) treatment × sex interaction was found for both weight gain and feed conversion efficiency. Therefore the results for both sexes were combined. Mean weight gain of the pigs was 715 g day^{-1} during the experimental period of 4 weeks. The results show that weight gain increased up to a dietary tryptophan level of 2.10 g kg^{-1}. With an increasing level of dietary tryptophan up to a content of 2.10 g kg^{-1} feed conversion improved.

The results of the IVVO trials (Table 6) agree well with those of the ILOB trials. Maximum weight gain was again established at a dietary tryptophan level of 2.10 g kg^{-1}. Maximum efficiency of feed utilization in these trials was found at a dietary level of 1.95 g kg^{-1} tryptophan. In the ILOB as well as in the IVVO trials there was a trend of a decreased performance at a dietary tryptophan level of 2.25 g kg^{-1}. These data suggest an imbalance caused by excess of tryptophan. Similar effects of a decreased performance when tryptophan was in excess of the requirement were reported by Lewis *et al.* (1977) and Borg *et al.* (1987).

Thus from the results of the current trials it can be concluded that the pig's requirement for total tryptophan in the weight range of 20 to 40 kg is approximately 2.1 g kg^{-1}. This value corresponds with 1.77 g kg^{-1} apparent ileal digestible tryptophan. The value of 2.1 g kg^{-1} is similar to those reported by Russel *et al.* (1983), Bertram & Berende (1983) and Schutte *et al.* (1988). Lower requirement values, however, are reported by Henry *et al.*, (1986) and NRC (Anonymous, 1988). Since requirement figures for amino acids based on total contents in the diet are affected by the ileal digestibility of the amino acid of concern, it is generally accepted that the levels of ileal digestible amino acids form a more satisfactory basis in diet for-

Table 6. Mean combined results of the three IVVO growth experiments for daily weight gain, feed conversion and daily feed intake.

Group	Tryptophan content (g kg^{-1})		Weight gain (g d^{-1})[1]	Feed conversion (kg feed kg^{-1} gain)[1]	Feed intake (g d^{-1})
	total[2]	ileal[3] digestible			
1	1.65	1.32	678^{a}	1.915^{a}	1,298
2	1.80	1.47	733^{ab}	1.885^{ab}	1,382
3	1.95	1.62	799^{bc}	1.778^{b}	1,421
4	2.10	1.77	826^{c}	1.797^{b}	1,484
5	2.25	1.92	795^{bc}	1.841^{ab}	1,463
LSD ($P < 0.05$)			*67.0*	*0.114*	

[1] Values within a column not having a common superscript differ significantly ($P < 0.05$).
[2] Analysed contents.
[3] Calculated using the digestibility coefficient for tryptophan determined in the digestibility trial.

mulation. (Sauer & Ozimek, 1986; Van Weerden, 1989; Lenis, 1992). Published data on the requirement for ileal digestible tryptophan in pigs, however, are scarce. Lenis (1992) concluded from a review of the literature that the requirement for ileal digestible tryptophan of pigs for the live weight period of 23 to 45 kg is approximately 1.8 g kg^{-1}. This value is in close agreement with that estimated from the results of the present trials.

In literature the requirement for tryptophan and other essential amino acids is often expressed as a percentage of the dietary level of lysine. However, expressing requirement figures of amino acids relative to lysine is only of practical relevance when they are based on a level of lysine, which is required by the pig. The results of a recent study of Schutte & De Jong (1993) pointed out that the requirement for ileal digestible lysine is approximately 9.0 g kg^{-1} for pigs in the live weight period of 20 to 40 kg. An almost similar requirement figure for ileal digestible lysine of 8.8 g kg^{-1} for this live weight period was reported by Lenis (1992). Based on the analysed content of lysine in the basal diet and the results from the digestibility study, it can be calculated that the dietary level of ileal digestible lysine in the present study amounted to 9.7 g kg^{-1}. This means that the dietary level of ileal digestible lysine in the diets of the present study was in excess of the requirement. Relative to the requirement figure for ileal digestible lysine reported by Schutte & De Jong (1993), our estimated requirement figure for ileal digestible tryptophan is approximately 20%. The same value was reported by Lenis (1992) on an ileal digestible basis.

It should be noted that the requirement figure for ileal digestible tryptophan found in the present study, and that reported by Lenis (1992) is only true for the pig breeds used in Dutch practice. For other breeds an other requirement figure for ileal digestible tryptophan may be found, since the requirement for amino acids depend on the pig's capacity for protein deposition. However, data on body composition of young pigs are missing to support further discussion on this subject.

In addition to lysine, methionine and threonine, tryptophan is also available commercially. Supplementation of a diet with these amino acids therefore provides a mean for increasing the efficiency of utilization of dietary protein. As a result N excretion by pigs will be reduced. This was confirmed in a study reported by Schutte *et al.* (1993). Their study pointed out that the dietary level of protein in a practical diet for pigs in the weight range of 20 to 40 kg can be reduced with approximately 20 g kg^{-1} (from 178 to 156 g kg^{-1}), provided the diet with 156 g kg^{-1} of protein was adequately supplied with lysine, methionine, threonine and tryptophan. A calculation carried out by Schutte *et al.* (1993) indicated that such a decrease of the dietary protein level will reduce N excretion with approximately 20%.

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