

## **The effect of dietary crude protein concentration on growth performance, carcass composition and nitrogen excretion in entire grower-finisher pigs**

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Two experiments, a performance experiment ( $n = 72$ ) and a nitrogen balance ( $n = 16$ ) experiment were conducted to evaluate the effects of dietary crude protein (CP) concentration on growth performance, carcass characteristics and nitrogen excretion of pigs. Dietary CP concentrations in experimental diets (g/kg) were 207.5, 170, 150 and 122.5 for treatments 1, 2, 3 and 4, respectively, and were offered to individually-fed entire-male grower-finisher pigs (45 to 95 kg). The diets were formulated to contain 13.7 MJ digestible energy and 11 g total lysine/per kg. Synthetic lysine, methionine, threonine and tryptophan were added to achieve ideal protein status. There was a linear increase in food intake as CP concentration decreased ( $P < 0.05$ ). There was a quadratic response in daily live-weight gain and food conversion ratio ( $P < 0.05$ ) to the change in CP concentration ( $P < 0.05$ ), with an improvement in daily gain and food conversion ratio occurring as CP concentration declined to 150 g/kg and a deterioration in these parameters thereafter. There was a linear decrease ( $P < 0.05$ ) in lean meat proportion as CP concentration decreased. There was a linear decrease in urinary output ( $P < 0.05$ ), urinary pH ( $P < 0.01$ ) and slurry pH ( $P < 0.05$ ) as dietary CP concentration decreased. There was a quadratic response in urinary nitrogen output ( $P < 0.05$ ), total nitrogen output ( $P < 0.05$ ) and N utilization as dietary CP decreased. In conclusion, a dietary CP level of 150 g/kg was optimal in terms of growth performance and reduced nitrogen excretion.

*Keywords:* Crude protein; excretion; nitrogen; pigs

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### Introduction

Pig farming, in common with all forms of agriculture, has the potential to pollute the natural environment (Bateman, 1998). From the mid 1980's onwards, environmental concerns have arisen relating to animal production, with manure disposal and odour control particularly important in relation to swine production in many countries.

Owing to the feeding value and price of raw ingredients commonly used in the diets fed to pigs, least cost formulation is commonly used to produce the most economical diet, which consequently leads to oversupply of certain amino acids and of protein generally (Gatel and Grosjean, 1992). Lenis (1989) remarked that non-essential amino acids are always supplied in adequate or excessive amounts when diets are formulated from natural feed-stuffs that provide the required essential amino acids. In addition to the substantial N excretion associated with high crude protein diets, work by Chen *et al.* (1999) has demonstrated reduced animal performance where excessive dietary crude protein levels were fed to growing pigs due to the metabolic cost of increased N excretion. Nitrogen excretion can be reduced substantially by supplying dietary amino acids in close accordance with the animal's requirement and by the incorporation of more free amino acids in feeds and lowering crude protein concentration (van Klooster *et al.*, 1998). The potential to maximise the reduction in nitrogen excretion without loss of animal performance, is dependent on maintaining nitrogen retention as dietary crude protein is reduced (Lee *et al.*, 1993).

Numerous researchers have studied the effects on animal performance and N excretion resulting from the lowering of dietary crude protein concentration accompanied by the addition of synthetic

amino acids (Lenis, 1989; Kephart and Sheritt, 1990; Gatel and Grosjean, 1992; Kerr and Easter, 1995; Tuitoek *et al.*, 1997; Canh *et al.*, 1998). The above authors reported substantial reductions in N excretion with reduced crude protein concentration, but variable results in relation to growth and carcass performance dependant on the magnitude of crude protein reduction and the extent of amino acid addition. Studies in which dietary crude protein level has been dropped below 130 g/kg have predominately involved castrate males and gilts (Kerr and Easter, 1995; Canh *et al.*, 1998). The objectives of the current experiment were (i) to compare the effect of dietary crude protein concentration on growth performance and carcass characteristics in entire male grower-finisher pigs, and (ii) to determine the effects of dietary crude protein level on nitrogen intake and excretion.

### Materials and Methods

#### *Animals and diets*

Two experiments were conducted, a performance trial and a nitrogen balance. The animals used in both experiments were Large White × (Large White × Landrace) entire males from the same genetic and health-status source. The experimental diets used in both experiments contained dietary crude protein concentrations (g/kg) as follows: 207.5 (T1), 170 (T2), 150 (T3) and 122.5 (T4). Diets were formulated using the feeding values for the ingredients as specified by R&H Hall, Technical Bulletin (1996). All treatments were formulated to contain 13.7 MJ digestible energy (DE) per kg and a total lysine concentration of 11 g/kg and were in pelleted form. Diets were supplemented with synthetic lysine, methionine, threonine and tryptophan so as to achieve ideal protein status as proposed

by Close (1994). Diet composition and nutrient analysis are detailed in Table 1.

#### *Performance study*

*Experimental design and management:* The building used had 12 pens, each of which could accommodate six pigs. Each pen was equipped with feeding crates to facilitate individual feeding. The pens consisted of a 70:30 solid:slatted floor area. The house was mechanically ventilated to provide an ambient temperature of 19 °C. The pigs had a space allowance of 0.75 m<sup>2</sup>/pig and were allowed into individual feeding crates at feeding time.

The experiment was designed as a complete randomised block design comprising four dietary treatments. Seventy-two pigs were used (initial mean live weight of 44.6

kg (s.d. 1.2) and were approximately 14 weeks of age). The pigs were blocked on the basis of live weight and allocated at random to the four dietary treatments. Pigs were grouped in pens of six, with all pigs within a pen being offered the same dietary treatment. The pigs were allowed 7 days to become familiar with the individual feeding crates and wet feeding and were fed twice daily on a scale ranging from 1.5 kg per day at the beginning to *ad-libitum* feeding. The individual feeding troughs were checked daily after the evening feed and food intake was recorded. If the pig had consumed all its allocated food, its food allowance was increased by 100 g for the following day, up to *ad-libitum* food intake. The food was presented as a pellet mixed with water in the

**Table 1. Ingredients and chemical composition of experimental diets (g/kg)**

	Diet			
	T1	T2	T3	T4
<i>Ingredients (g/kg)</i>				
Wheat	462.4	558.7	628.4	770
Soyabean meal (Hi-pro)	235.6	129.5	51.9	0
Peas	100	100	100	0
Pollard	150	150	150	150
Tallow	28.7	31.7	33.8	37.5
Salt	5	5	5	5
Minerals and vitamins	2.5	2.5	2.5	2.5
Dicalcium phosphate	1.62	3.4	4.7	5.8
Limestone	14.2	13.6	13.1	12.8
Synthetic lysine	0	3.7	6.34	9.65
Synthetic methionine	0	0.5	1.3	1.9
Synthetic threonine	0	1.45	2.7	4.1
Synthetic tryptophan	0	0	0.31	0.7
<i>Chemical composition (g/kg)</i>				
Dry matter	879.8	880.6	881.9	876.5
Ash	48.2	45.2	43.8	36.7
Ether extract	51.2	55.3	57.8	55.6
Neutral detergent fibre	157.2	156.2	148.2	164.4
Crude protein	207.5	170.0	150.0	122.5
Gross energy (MJ/kg)	16.7	16.7	16.7	16.6
Lysine	11.52	11.41	11.26	11.16
Methionine and cystine	7.12	6.47	6.39	6.37
Threonine	7.61	7.48	7.34	7.31
Tryptophan	2.24	2.12	1.94	1.91

proportion 1:2 w/v. Pigs had access to another source of water in the pen at all times. The pigs were allowed a minimum of 1 h and maximum of 1 h 30 min feeding time, morning and evening. Animals were weighed at the start of the experiment and subsequently at 2-week intervals through to slaughter.

*Blood sampling:* The pigs were blood sampled on day 7, 21, 35 and 49, 2 h after the cessation of morning feeding, to determine serum urea levels. Blood samples were collected from the cephalic vein into 10-ml vacutainer tubes. Samples were stored for 24 h at 4 °C and then centrifuged at 2500 rpm for 10 min. Serum was then separated and stored frozen until analysed.

*Carcass analysis:* Slaughter was in two batches after individual live weights exceeded 96 kg. Hot carcass weight (HCW) was recorded approximately 1 h post-mortem. Subcutaneous backfat measurements (P2) were determined using an introscope positioned 6.5 cm from the midline split of the carcass at the last rib. Further carcass traits were determined by application of the following equations:

$$\text{Carcass weight (CW) (kg)} = \text{HCW} \times 0.98$$

$$\begin{aligned} \text{Lean meat proportion (g/kg)} \\ &= 655 - (11.5 \times \text{P2}) + 0.76 \text{ CW} \\ &\text{(Whittemore, 1993)} \end{aligned}$$

$$\begin{aligned} \text{Kill-out proportion (g/kg)} \\ &= \text{CW/slaughter weight} \end{aligned}$$

$$\begin{aligned} \text{Carcass daily gain (kg/day)} \\ &= (\text{CW} - (\text{initial live weight} \times \\ &0.65))/\text{days on experiment} \end{aligned}$$

$$\begin{aligned} \text{Carcass FCR} &= \text{total food intake}/ \\ &(\text{CW} - (\text{initial live weight} \times 0.65)) \end{aligned}$$

#### *Nitrogen balance study*

Sixteen pigs were used in this study (initial weight 53 kg (s.d. 0.6)). Prior to their

selection these animals were group housed and fed a commercial finisher ration. For the purpose of the nitrogen balance trial, the animals were housed in a climate-controlled house with temperature maintained at 20 °C. The pigs were blocked on the basis of live weight and allocated at random to the four dietary treatments and housed in metabolism crates. The metabolism crates allowed for the collection of urine and faeces separately. Pigs were allowed a 5-day acclimatisation period, followed by a 7-day collection period. Food and water were offered *ad-libitum* and quantities consumed were recorded. Water was supplied along with the food and between feeds as required. Urine was collected daily in buckets acidified by the addition of 20 ml H<sub>2</sub>SO<sub>4</sub> (25% v/v) so as to minimise the atmospheric loss of nitrogen. Urine chutes were flushed with 10 ml of H<sub>2</sub>SO<sub>4</sub> (2% v/v) daily to recover any urine adhering to the surface. Samples of fresh faeces were taken daily and frozen for subsequent analysis. Urinary, faecal and slurry pH were measured over 2 days following the nitrogen balance collection period, during which time acid was not added to the urine bucket to facilitate an accurate pH reading for the urine. Determination of faeces pH was facilitated by mixing a fresh faeces sample with distilled water (v/w). The daily quantities of urine and faeces were mixed during these 2 days to form a slurry from which a pH reading was determined daily.

#### *Laboratory analyses*

The determination of dietary dry matter (DM), crude protein (CP) and ash were carried out according to the Association of Official Analytical Chemists (AOAC) (1980). Diets were hammer milled through a 1-mm screen (Christy and Norris, Chelmsford, England) prior to analysis. The dietary DM concentration

was determined following oven drying at 55 °C for 72 h. Faecal DM matter was determined following drying at 100 °C for 72 h. Dietary and faecal ash concentrations were determined following combustion in a furnace at 600 °C for 4 h. Dietary crude protein concentration and urinary nitrogen concentration were determined as Kjeldahl N  $\times$  6.25 using the LECO FP 528 instrument (Leco Instruments, UK Ltd, Cheshire). The nitrogen concentration of the fresh faeces was determined by the macro-Kjeldahl method (Kjeldahl N  $\times$  6.25) (AOAC, 1980) using a Buchi 323 distillation unit (Buchi, Switzerland). Neutral detergent fibre (NDF) was determined by the method of Van Soest (1976), ether extract (EE) was determined using the 1043 Soxtec System HT6 as derived from the Soxhlet method. Dietary and faecal gross energy were determined using an adiabatic bomb calorimeter (Parr, Illinois, USA). Dietary amino acid concentrations were determined by the method of Iwaki *et al.* (1987). Urea nitrogen was determined by the Urease-Berthelot method, using an enzymatic kit

(Randox Laboratories Ltd, Co. Antrim, N. Ireland).

#### Statistical analysis

The experimental data were analysed using the General Linear Models Procedure (PROC GLM) of the SAS (SAS Institute, 1985). The statistical model included effects for treatment, block and block-by-treatment. Linear and quadratic effects of dietary CP concentration were evaluated. The individual pig was the experimental unit in all analyses. Performance data were adjusted for initial live weight by covariance. Slaughter weight was used as the covariate for adjustment of carcass data. Blood urea data were analysed using a repeated measures analysis using PROC MIXED of SAS (Littel *et al.*, 1996).

## Results

### Performance study

The effects of dietary treatment on live weight, food intake, average daily live-weight gain (ADG) and food conversion

**Table 2. Effect of dietary protein concentration on food intake, growth rate and food conversion ratio (least squares means) (Performance study)**

Trait	Dietary protein concentration (g/kg)				s.e.	Contrasts	
	207.5 (T1)	170.0 (T2)	150 (T3)	122.5 (T4)		Linear	Quadratic
<i>Daily gain (g/day)</i>							
Grower (day 0 to 28)	815	823	897	816	23.0	*	*
Finisher (day 28 to slaughter)	930	948	1022	956	31.0		
Overall	858	875	944	882	21.0		*
<i>Food intake (kg/day)</i>							
Grower period (day 0 to 28)	1.75	1.75	1.94	1.92	0.041	**	
Finisher (day 28 to slaughter)	2.47	2.35	2.56	2.53	0.062		
Overall	2.11	2.04	2.22	2.23	0.049	*	
<i>Food conversion ratio (kg/kg)</i>							
Grower (day 0 to 28)	2.16	2.15	2.19	2.39	0.055	*	*
Finisher (day 28 to slaughter)	2.70	2.49	2.53	2.74	0.085	*	*
Overall (day 0 to slaughter)	2.47	2.35	2.36	2.57	0.053	**	*

ratio (FCR) are presented in Table 2. Performance data were analysed for the following periods; days 0 to 28 (grower period), day 28 to slaughter (finisher period) and day 0 to slaughter (grower-finisher). There was a linear increase in food intake with declining crude protein concentration during the grower period ( $P < 0.01$ ) and during the grower-finisher period ( $P < 0.05$ ). There was a quadratic response in daily live-weight gain ( $P < 0.05$ ) to the decreasing crude protein concentration during the grower period and during the grower-finisher period. There was a quadratic response ( $P < 0.05$ ) in FCR to the decreasing crude protein concentration during the grower, finisher and grower-finisher periods.

The effects of dietary treatment on carcass traits are presented in Table 3. There was a linear decrease ( $P < 0.05$ ) in lean meat proportion and a linear

increase ( $P < 0.05$ ) in back fat depth to decreasing crude protein concentration. There was a quadratic response in carcass daily gain ( $P < 0.05$ ) and carcass FCR ( $P < 0.01$ ) to the decreasing crude protein concentration. Serum urea nitrogen concentrations (Table 4) decreased ( $P < 0.05$ ) as dietary crude protein concentration decreased.

#### Nitrogen balance study

The effects of dietary treatment on nitrogen balance are presented in Table 5. There was a linear decrease in nitrogen intake ( $P < 0.05$ ) and water intake ( $P < 0.05$ ) as dietary crude protein concentration decreased. There was a linear decrease in urinary output ( $P < 0.05$ ), slurry output, urinary pH ( $P < 0.01$ ) and slurry pH ( $P < 0.05$ ) as dietary crude protein concentration decreased. There was a quadratic response in urinary nitrogen

**Table 3. Least squares means for weight at slaughter and carcass traits (Performance study)**

Trait	Dietary protein concentration (g/kg)				s.e.	Contrasts	
	207.5 (T1)	170.0 (T2)	150 (T3)	122.5 (T4)		Linear	Quadratic
Slaughter weight (kg)	95.2	96.5	95.7	95.4	1.47		
Carcass weight (kg)	70.7	71.0	71.3	71.1	0.41		
Kill-out proportion (g/kg)	739.1	741.1	745.0	743.1	4.15		
Lean proportion (g/kg)	580.5	581.4	566.4	565.1	6.93	*	
Backfat (mm)	10.74	10.66	11.96	12.08	0.603	*	
Carcass gain (kg/day)	0.710	0.717	0.792	0.724	0.019	*	*
Carcass FCR (kg/kg)	2.99	2.87	2.82	3.09	0.062	*	**

**Table 4. Least squares means for the effect of dietary protein concentration and time on serum urea nitrogen concentration (mg/dL) (Performance study)**

Time (days)	Dietary protein concentration (g/kg)				s.e.	Contrasts	
	207.5 (T1)	170.0 (T2)	150 (T3)	122.5 (T4)		Linear	Quadratic
7	32.2	24.2	20.1	14.1	0.87	*	
21	33.3	23.6	17.4	16.3	0.85	*	*
35	33.5	24.5	21.1	16.2	0.88	*	
49	33.3	23.2	20.4	13.3	0.88	*	
Overall	33.1	23.9	19.7	15.0	0.99	*	

**Table 5. Effect of dietary treatment on nitrogen balance, nutrient digestibility coefficients and slurry characteristics (least squares means) (Nitrogen balance study)**

Trait	Dietary protein concentration (g/kg)				s.e.	Contrasts	
	207.5 (T1)	170.0 (T2)	150 (T3)	122.5 (T4)		Linear	Quadratic
Food intake (kg/day)	2.14	2.26	2.31	2.53	0.079		
Water intake (kg/day)	6.25	6.01	5.56	5.53	0.11	*	
Nitrogen intake (g/day)	71.2	61.5	55.6	49.5	8.00	*	
Urine output (litres/day)	3.50	3.04	2.70	2.59	0.334	*	
Faecal output (kg/day)	1.02	1.17	1.05	1.14	0.107		
Slurry output (kg/day)	4.52	4.20	3.76	3.73	0.355	*	
Urine pH	8.67	8.60	7.64	6.11	0.326	**	
Slurry pH	7.56	7.62	7.31	6.68	0.238	*	
Urinary nitrogen (g/day)	26.5	18.8	16.5	13.8	2.92		*
Faecal nitrogen (g/day)	9.4	9.9	7.4	8.9	1.08		
Total nitrogen excretion (g/day)	35.9	28.7	23.9	22.7	0.83		*
Nitrogen utilisation (N retained/N intake)	0.49	0.53	0.57	0.54	0.002		*
Nitrogen digestibility (g/g)	0.869	0.837	0.868	0.841	0.002		
Dry matter digestibility (g/g)	0.857	0.856	0.862	0.854	0.006		
Gross energy digestibility (MJ/MJ)	0.858	0.858	0.863	0.854	0.005		
Digestible energy concentration (MJ/kg)	14.3	14.3	14.4	14.1	0.50		

output ( $P < 0.05$ ), total nitrogen output ( $P < 0.05$ ) and nitrogen utilisation as dietary crude protein concentration decreased.

### Discussion

Diets formulated to reduce nitrogen excretion by pigs will only be acceptable to the pig industry if they maintain pig performance (Kay and Lee, 1996). Feeding of high protein diets has been shown to limit voluntary feed intake in growing pigs (Henry, 1985; Forbes, 1995). In the present study, there was a linear increase in feed intake to decreasing crude protein concentration. An excessive protein concentration in food leads to increased heat production from deamination of the excess amino acids, which may depress intake if heat dissipation becomes limiting and body temperature rises or if the products of deamination become marginally toxic (Forbes, 1995).

In agreement with the current performance study, Campbell (1988) and Chen *et al.* (1999) demonstrated deterioration in ADG and FCR as CP level increased above the optimal level for the particular sex and genotype. The negative effects of an increased protein level in the diet, for a given lysine concentration, on live-weight gain and FCR is primarily due to extra heat increment originating from the catabolism of excess amino acids (Henry, Colleaux and Seve, 1992). Part of the growth decreasing effect of high CP diets may be due to the reduced net energy value of the diet as a result of reduced utilisation efficiency of metabolisable energy due to amino acid catabolism (Hansen and Lewis, 1993). This catabolic penalty of an increasing protein turnover should inevitably reduce the biological value of the dietary protein (Roth *et al.*, 1999).

Conversely, the deterioration in performance traits noted as crude protein

concentration was reduced further to 122.5 g/kg is in line with the work of Kephart and Sherritt (1990) who found that the addition of glutamic acid to a 12% crude protein diet, balanced for the first four limiting amino acids failed to achieve similar N utilisation as a diet of 16% crude protein, indicating that another essential amino acid, or a combination of essential and non-essential amino acids may be limiting. Kerr and Easter (1995) noted that the addition of non-essential amino acids resulted in improved N utilisation in low protein diets. The diets used in the current experiment were analysed for lysine, methionine, threonine and tryptophan concentration and met the ideal protein requirements given by Wang and Fuller (1989). However, the remaining essential and non-essential amino acid concentrations were not determined. It is likely that the deterioration in live-weight gain and FCR with the diet of 122.5 g/kg CP may be a result of either a deficiency of some amino acid, a deficiency of N generally or a combination of both.

In agreement with the findings of Chen *et al.* (1999) and Gomez *et al.* (2002), the serum urea concentration declined as CP concentration declined, indicating a reduction in excess dietary nitrogen intake. Those pigs fed 122.5 g/kg CP had serum urea levels which on average were proportionately 0.55, 0.37 and 0.24 lower than those fed CP levels of 207.5, 170 and 150 g/kg reflecting a reduction in excess dietary N. Work by Chen, Miller and Lewis (1994a,b) and Coma, Zimmerman and Carrion (1995) has shown that fluctuations in plasma urea concentration over time can be used to determine the protein and amino acid requirements of the growing pig, with a decline in plasma urea nitrogen indicating an approach to the animals requirements.

The effect of reduced CP concentration on carcass fatness is in agreement with

Kay and Lee (1996) who reported an increased level of backfat deposition in grower-finisher pigs as dietary protein level is reduced. Van Lunen and Cole (2001) noted that the deamination and elimination of excess dietary protein is an energy inefficient process and therefore less energy is available in high-CP diets for fat synthesis. Although not measured in the current experiment, the reduced deamination cost associated with the lower-CP diets may have resulted in an increase in net energy, since Dourmad *et al.* (1993) found no difference in backfat depth where diets of low and high crude protein were formulated to contain similar net energy concentrations.

The results of the current study indicate proportional decreases of 0.34 and 0.37 in total daily N excretion as dietary crude protein level was reduced from 207.5 to 150 and 122.5 g/kg, respectively. This decrease in N excretion equates to a proportional reduction of 0.06 in N excretion per 1% reduction in crude protein level to 150 g/kg with no significant advantage in N excretion being gained from reducing crude protein level from 150 to 122.5 g/kg. Kerr and Easter (1995) concluded that for each one-percentage unit reduction in dietary crude protein combined with amino acid supplementation, total N excretion (faecal plus urinary) could be reduced by approximately 8%. The results of the current experiment show no significant reduction in N excretion below 150 g/kg CP which is in conflict with work by Dourmad *et al.* (1993) and Canh *et al.* (1998), who noted significant reductions in N excretion when crude protein concentration was reduced from 155 to 136 g/kg and 145 to 125 g/kg, respectively.

The reductions in urinary-N excretion in response to reduced dietary CP levels in the current experiment are in agreement with work by Quiniou, Dubois and Noblet

(1995), Carter *et al.* (1996) and Le Bellego *et al.* (2001). Jongbloed and Lenis (1992) noted that a much higher proportion of the pig's N excretion appears in the urine, mainly as a result of an oversupply and/or imbalance of amino acids which cannot be used for body protein deposition. A reduction in the non-essential N level in the diet is therefore associated with a decreased N loss in urine (Sharda, Mahan and Wilson, 1976).

In the current study, there was a linear decrease in slurry pH as dietary crude protein concentration decreased. Work by Canh *et al.* (1998) indicated similar significant reductions in slurry pH and consequently a reduction in ammonia volatilisation where dietary CP concentration was reduced.

In conclusion, the results of the current experiment indicate that altering the dietary CP concentration has significant effects on growth performance and nitrogen excretion in entire male grower-finisher pigs. In the case of the sex, genotype and environment present in the current study, daily gain was seen to improve as CP concentration was reduced from 207.5 g/kg to 150 g/kg and deteriorate thereafter. From an environmental protection point of view the current experiment confirms that reducing dietary crude protein levels can effectively reduce N excretion, although no significant advantage was to be gained from reducing crude protein level below 150 g/kg.

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Received 10 July 2003