

## Reducing crude protein content with supplementation of synthetic lysine and threonine in barley - rapeseed meal - pea diets for growing pigs

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This study was conducted to determine the possibility to use synthetic amino acids to lower the nitrogen output from pig production. A performance experiment was carried out with 120 triplet-fed growing pigs whose dietary crude protein was reduced from 179 g/feed unit (FU= 0.7 kg starch equivalent) to 160, 140 and 122 g/FU, respectively. The diets were supplemented with synthetic lysine and threonine to keep the level of these amino acids constant.

Dietary protein reduction did not affect the growth performance or feed conversion ratio of the pigs, but it did linearly increase the portion of fat to lean in the carcass. Significant linear effect was found in back fat ( $p < 0.001$ ) and side fat thickness ( $p < 0.01$ ) and in the proportion of lean in valuable cuts and in whole carcass ( $p < 0.05$ ).

However, the change in carcass composition was negligible down to a crude protein level of 140 g/FU. It may, therefore, be concluded that the crude protein content of the diet can be reduced by up to 20% by balancing the dietary protein with synthetic lysine and threonine.

Keywords: pigs, growth rate, feed conversion ratio, carcass composition, nitrogen excretion, crude protein, lysine, threonine

### Introduction

Animal production causes serious environmental problems in many European countries. The increasing content of nitrogen in the soil, atmosphere and fresh water, originating from manure, is a real problem in areas with high-density animal production (LENIS 1989, VERSTEGEN and TAMMIGA 1991).

Much effort has been focused on nutritional means of reducing environmental pollution (JONGBLOED and LENIS 1992). In pig production, the lowering of nitrogen excretion is related to the protein content and amino acid balance of the diet,

since the oversupply of protein and the imbalanced amino acids are excreted mostly in urine. A closer balance of amino acids in the diet compared to the requirement can be achieved, if feeding is based on the requirements set by age and physiological state, or if the availability and quality of the dietary protein are improved (LENIS 1989).

The availability of economically viable synthetic amino acids has made them an attractive alternative in balancing the dietary amino acid composition of pig feeds. A significant reduction in the crude protein content of cereal-based diets has been attained with the supplementation of synthetic lysine alone,

without impairing the performance of the pigs (TAYLOR et al. 1979, EASTER and BAKER 1980). Furthermore, the reduction in dietary protein has also depressed water consumption of the pigs, which again leads to a smaller volume of excreted urine (PFEIFFER and HENKEL 1991).

This experiment was undertaken to make a comparison between the present protein feeding recommendations for growing pigs in Finland based on the digestible crude protein (SALO et al. 1990) and balancing of essential amino acid content of the diet with synthetic amino acids with respect to the requirements of growing pigs. The object of the experiment was to determine the effects of the reduction in the crude protein content of the diet on the performance and carcass composition of growing pigs and on the nitrogen output from swine production.

## Material and methods

A total of 120 Landrace and Yorkshire pigs, weighing 25.5 kg (standard error 0.19) on average, were used in the performance trial. Three pigs of the same sex and different litter origin were placed into each pen and the pens were allotted at random to one of four dietary treatments. The treatment groups were balanced according to initial weight and sex. Each diet was tested on 15 female and 15 castrated pigs.

The crude protein contents in the four diets were 179, 160, 140 and 122 g/FU, respectively. The control diet, containing 179 g/FU crude protein, consisted of barley, rapeseed meal (*Brassica campestris*) and peas (var. Proco). The crude protein content of the three experimental diets was reduced, while the average lysine and threonine levels were maintained at 9.0 and 6.2 g/FU with synthetic lysine and threonine. Commercially produced lysine hydrochloride and threonine (Eurolysine) were used as supplementary amino acids. All diets contained an adequate amount of minerals and vitamins to meet the nutritional requirements of growing pigs (SALO et al. 1990). The composition of the four diets is presented in Table 1. The feed ingredients were analyzed by standard methods (AOAC

Table 1. Dietary ingredients (g/kg air-dry diet) and calculated chemical composition of the diets.

| Diet                                | 1     | 2     | 3     | 4     |
|-------------------------------------|-------|-------|-------|-------|
| Ingredients:                        |       |       |       |       |
| Barley                              | 575   | 665   | 755   | 845   |
| Rapeseed meal                       | 260   | 200   | 140   | 80    |
| Peas                                | 130   | 100   | 70    | 40    |
| Mineral + vitamin mix. <sup>2</sup> | 35    | 35    | 35    | 35    |
| Synthetic amino acids               |       |       |       |       |
| L-lysine HCl, g/kg                  | -     | 1.13  | 2.26  | 3.40  |
| Threonine, g/kg                     | -     | 0.51  | 1.02  | 1.55  |
| Chemical composition:               |       |       |       |       |
| Crude protein, g/kg                 | 163   | 148   | 132   | 117   |
| " , g/FU <sup>1</sup>               | 179   | 160   | 140   | 122   |
| Digestible crude protein, g/kg      | 128   | 117   | 105   | 93    |
| " , g/FU                            | 141   | 126   | 111   | 97    |
| Lysine, g/FU                        | 9.0   | 9.0   | 9.0   | 9.0   |
| Threonine, "                        | 6.4   | 6.3   | 6.1   | 5.9   |
| Methionine+Cystine, "               | 6.7   | 6.1   | 5.5   | 4.9   |
| Histidine, "                        | 5.1   | 4.8   | 3.8   | 3.2   |
| Leucine, "                          | 14.0  | 12.4  | 10.9  | 9.4   |
| Isoleucine, "                       | 7.9   | 6.9   | 6.0   | 5.1   |
| Phenylalanine, "                    | 8.9   | 8.1   | 7.2   | 6.4   |
| Arginine, "                         | 11.7  | 10.0  | 8.4   | 6.9   |
| Valine, "                           | 9.7   | 8.6   | 7.6   | 6.6   |
| Feed unit (FU/kg DM)                | 1.04  | 1.06  | 1.07  | 1.09  |
| ME, MJ/kg                           | 14.30 | 14.41 | 14.51 | 14.62 |

<sup>1</sup> Feed unit (FU)= 0.7 kg starch equivalent.

<sup>2</sup> Composition of mineral and vitamin mixture (per kg): Ca 200 g, P 75 g, Mg 11.5 g, NaCl 103 g, Fe 3000 mg, Zn 2674 mg, Mn 1151 mg, Cu 1000 mg, Co 38 mg, J 6 mg, Se 2.8 mg, vitamin A 350 000 IU, vitamin D<sub>3</sub> 57 200 IU, vitamin E 1200 mg, thiamine 57 mg, riboflavine 200 mg, pyridoxine 114 mg, pantothenic acid 430 mg, folic acid 114 mg, B<sub>12</sub>-vitamin 570 µg and biotin 4000 µg.

1984), and their amino acid composition was determined by gas-liquid chromatography (GLC).

The pigs were housed in partially slatted pens with concrete floors and had free access to water. Feeds were given on a restricted scale in relation to live weight twice daily (1.2-3.0 FU/day) according to Finnish feeding standards (SALO et al. 1990). Daily allowance was increased by 0.2 FU/animal/week in the beginning of the experiment and by 0.1 FU/animal/week after the ninth experimental week. The pigs were slaughtered at an average weight of 105 kg.

The carcass weight was recorded 24 h after slaughter. The left half of the cold carcass of each

pig was cut into valuable cuts (ham, fore-end, shoulder and loin), and the cuts were dissected to determine lean (including bones) and fat (including skin). The proportion of lean and fat was calculated from the ham, valuable cuts and whole carcass. The area of the *longissimus dorsi* muscle was determined posterior from the last rib with a planimeter. The thickness of back fat was calculated as a mean of five measurements (shoulder, middle and ham (mean of three measurements)), and the thickness of side fat was determined from the end of *longissimus dorsi*.

Nitrogen excretion was calculated as the difference between nitrogen supply from the diet and total nitrogen retention during the experiment. The quantity of nitrogen retained in the body during the experiment was calculated as the difference between the nitrogen content of the pig at the end of the experiment and the estimated nitrogen content of the pig in the beginning of the experiment. The nitrogen retained in the carcass was estimated from the dissection results as a sum of the nitrogen in the carcass lean and bones (including head). The proportion of blood, intestines and offals was estimated as the difference between slaughter loss proportion and gut fill. The content of nitrogen in carcass lean and bones and the initial nitrogen content of the pigs were adapted from the dissection and the chemical analysis results of 40 pigs slaughtered at regular intervals between 20 and 110 kg live weight (ALAVIUHKOLA et al., unpublished). The proportion of gut fill and the content of nitrogen in the blood, intestines and offals were estimated from the results of WHITTEMORE and ELSLEY (1976).

The data were analyzed by the GLM procedure of SAS (1990). The hierarchic analysis of variance was performed as outlined by GILL (1989) using the model:  $y_{ijkl} = \mu + d_i + s_j + (ds)_{ij} + p_k(ds)_{ij} + e_{ijk}$ , where  $d$ ,  $s$  and  $p$  are diet, sex and pen effects, respectively,  $ds$  is the effect of diet\*sex, and  $e_{ijk}$  is the error term. The effect of pen within diet\*sex interaction ( $p_k(ds)_{ij}$ ) was used as an error term for testing the diet and sex effects. Feed consumption was registered on pen basis and the pen effect was, therefore, excluded from the model in statistical testing. Slaughter weight was used as a covariate in

the testing of carcass trait measurements. The degrees of freedom for the diet effect were further partitioned into single degrees of freedom to test the linear, quadratic and cubic trends of the crude protein level.

## Results

One gilt on diet 4 was excluded from the trial for a reason unrelated to the treatment. Otherwise the pigs completed the experiment successfully, and refusals of the diets were negligible. The overall results of the experiment are given in Table 2. The quadratic and cubic trends of the crude protein content and diet\*sex interaction were non-significant and are therefore omitted from Table 2.

## Performance

The growth performance of the pigs was good over the entire experimental period, averaging 880 g/day. The daily gain (DG) and feed conversion ratio (FCR) of the pigs were unaffected by the reduction in the dietary crude protein level. Total ( $p < 0.05$ ) and daily ( $p < 0.01$ ) feed consumption (measured as feed units) increased linearly with the reduction in the crude protein content. No significant differences between sexes were found in the performance of the pigs.

## Carcass composition

There was a significant difference in carcass weight ( $p < 0.05$ ) between the four groups, which also produced a difference in the corrected final weight ( $p < 0.05$ ). The carcass composition of the pigs changed from lean to fat when the crude protein content in the diet decreased, while a significant linear increase was found in back fat ( $p < 0.001$ ) and side fat ( $p < 0.05$ ) thickness. The lean content in valuable cuts and in the carcass also decreased linearly ( $p < 0.05$ ). However, no significant differences were found in the area of the *longissimus dorsi* muscle between the treatments.

Table 2. Performance and carcass composition of the pigs (LS means of the treatments are presented).

| Treatment                                 | 1     | 2     | 3     | 4     | SEM   | Sex     |                 | SEM   | Significance   |     |
|---|-------|-------|-------|-------|-------|---------|-----------------|-------|----------------|-----|
|   |       |       |       |       |       | Females | Castrated males |       | treatm. linear | Sex |
| Crude protein, g/FU                       | 179   | 160   | 140   | 122   |       |         |                 |       |                |     |
| No. of animals                            | 30    | 30    | 30    | 29    |       | 59      | 60              |       |                |     |
| Initial weight, kg                        | 25.4  | 25.5  | 25.5  | 25.5  | 0.32  | 25.7    | 25.2            | 0.22  | NS             | NS  |
| Final weight, kg                          | 104.8 | 104.7 | 105.1 | 105.6 | 0.53  | 104.9   | 105.2           | 0.38  | NS             | NS  |
| Final weight (corr.), kg <sup>1</sup>     | 103.9 | 104.6 | 105.4 | 106.4 | 0.81  | 105.3   | 104.9           | 0.57  | *              | NS  |
| Daily gain, g                             | 871   | 881   | 890   | 883   | 11.1  | 881     | 881             | 7.84  | NS             | NS  |
| Days in exp.                              | 90.4  | 90.4  | 90.2  | 92.1  | 1.36  | 90.7    | 90.9            | 0.96  | NS             | NS  |
| Feed consumption, FU/animal               | 209.8 | 209.8 | 211.8 | 221.6 | 3.70  | 212.5   | 214.0           | 2.59  | *              | NS  |
| FU/day/animal                             | 2.32  | 2.32  | 2.35  | 2.41  | 0.022 | 2.34    | 2.35            | 0.015 | **             | NS  |
| FCR, kg DM/kg gain                        | 2.57  | 2.51  | 2.47  | 2.49  | 0.035 | 2.50    | 2.52            | 0.024 | o              | NS  |
| " , FU/kg gain                            | 2.67  | 2.65  | 2.65  | 2.74  | 0.037 | 2.67    | 2.69            | 0.026 | NS             | NS  |
| " , MJ ME/kg gain                         | 41.2  | 41.4  | 41.1  | 42.0  | 0.57  | 41.3    | 41.5            | 0.40  | NS             | NS  |
| Carcass weight, kg                        | 76.9  | 77.4  | 78.0  | 78.7  | 0.60  | 77.9    | 77.6            | 0.73  | *              | NS  |
| Loss at slaughter, %                      | 26.3  | 25.9  | 25.9  | 25.9  | 0.20  | 25.8    | 26.2            | 0.14  | NS             | NS  |
| Back fat thickness, mm                    | 24.4  | 25.0  | 26.3  | 27.3  | 0.51  | 25.3    | 26.2            | 0.37  | ***            | o   |
| Side fat thickness, mm                    | 16.5  | 17.7  | 17.9  | 18.9  | 0.55  | 16.5    | 19.0            | 0.39  | **             | *** |
| Area of <i>l. dorsi</i> , cm <sup>2</sup> | 43.4  | 43.0  | 42.7  | 43.3  | 0.59  | 44.7    | 41.4            | 0.42  | NS             | *** |
| Lean in ham, %                            | 81.8  | 81.6  | 81.0  | 80.7  | 0.43  | 81.7    | 80.8            | 0.30  | o              | *   |
| Lean in valuable cuts, %                  | 80.6  | 80.4  | 79.8  | 79.2  | 0.40  | 80.6    | 79.4            | 0.28  | *              | **  |
| Carcass lean, %                           | 53.9  | 53.7  | 53.0  | 52.9  | 0.37  | 53.9    | 52.9            | 0.26  | *              | **  |

Significance: NS= non-significant, o=p<0.10, \*= p<0.05, \*\*=p<0.01, \*\*\*=p<0.001. SEM=standard error of means. FCR= feed conversion ratio.

<sup>1</sup> Final weight corrected with 26.0 % loss at slaughter.

Significant differences between sexes were found in side fat thickness (p<0.001) and in the area of the *longissimus dorsi* muscle (p<0.001) as well as in the proportion of lean in ham (p<0.05), valuable cuts (p<0.01) and whole carcass (p<0.01). In all these measurements, gilts were found to be superior to castrated males.

### Nitrogen excretion

The total quantity of nitrogen retained in the body for treatments 1, 2, 3 and 4 was 1.65, 1.65, 1.64 and 1.64 kg/pig, respectively (Table 3). Correspondingly, the quantity of nitrogen ingested per pig was 6.01, 5.37, 4.74 and 4.33 kg, and nitrogen excreted (ingested - retained) was 4.36, 3.72, 3.10 and 2.69 kg, respectively.

Table 3. Calculated nitrogen balance of the pigs.

| Treatment  | 1    | 2    | 3    | 4    |
|--|------|------|------|------|
| Crude protein, g/FU  | 179  | 160  | 140  | 122  |
| Nitrogen ingested, kg/pig                                  | 6.01 | 5.37 | 4.74 | 4.33 |
| Nitrogen retained, kg/pig                                  | 1.65 | 1.65 | 1.64 | 1.64 |
| Nitrogen excreted, kg/pig                                  | 4.36 | 3.72 | 3.10 | 2.69 |
| Difference in nitrogen excretion compared to control group |      |      |      |      |
| kg/pig   | -    | 0.63 | 1.24 | 1.67 |
| %  | -    | 14.7 | 28.4 | 38.3 |

### Discussion

The results of this study clearly confirmed that it was possible to reduce nitrogen excretion in swine production by applying a suitable feeding strategy.

In our experiment, lysine and threonine were chosen as the supplementary amino acids because they are the two most limiting amino acids in barley-based diets (FULLER et al. 1979a, 1979b). Similar results have been also obtained in other performance trials (TAYLOR et al. 1979, EASTER and BAKER 1980) and balance trials (GATEL and GROSJEAN 1992, NÄSI 1985) with the supplementation of lysine only. EASTER and BAKER (1980) reduced the dietary crude protein content of a corn-soybean meal diet by 10% without observing any negative effects on the performance of the pigs. Furthermore, TAYLOR et al. (1979) showed that the crude protein content of a barley-soybean meal diet could be lowered from 17.6 to 14.5% , but at lower levels the performance of the pigs started to deteriorate. GATEL and GROSJEAN (1992) reported that a 7.5% decrease in nitrogen intake resulted in a 15 to 20% decrease in nitrogen excretion in a balance trial with growing pigs. In another trial by the same authors, in which soybean meal was substituted by peas and supplemented with synthetic lysine, threonine, methionine and tryptophan, even a 35% decrease in nitrogen excretion could be achieved by a 15% decrease in nitrogen intake. NÄSI (1985) found that there was not any further increase in nitrogen retention after lysine supplementation when methionine was added to a low-protein diet of growing pigs.

A progressive reduction in crude protein with the supplementation of only one amino acid leads to a lack of other essential amino acids or non-essential protein. In our experiment, the deterioration in carcass trait measurements indicated that amino acids other than lysine or threonine or non-essential protein became limiting factors for lean growth. Histidine is regarded as the third limiting amino acid in barley (FULLER et al. 1979a), but its content in diet 4 was found to be on the level of the minimum recommendation of ARC (1981), 3 g/kg vs 2.8 - 4.0 g/kg. The content of sulphur-containing amino acids may also have been a limiting factor for lean growth in treatment 4 when the balance between essential amino acids was compared to ideal protein determined by WANG and FULLER (1989) (54 vs 63). The ratio of threonine to lysine in diet four was lower than that reported by WANG and FULLER

(1989) (66 vs 72), but it was above the ratio given by ARC (1981) (60). On the other hand the calculations of ARC (1981) and the present study are based on a total amino acid basis, whereas WANG and FULLER (1989) used ileal digestible amino acids in their determination. The ratio between essential and non-essential amino acids in all diets was above the minimum ratio (45:55) of WANG and FULLER (1989).

TAYLOR et al. (1979) reported, that the deterioration of carcass quality started at a higher level of dietary protein compared to the deterioration in the performance of the pigs. The fact that the highest content of lean was found in the control group could also be the result of using excess dietary protein for energy, since crude protein is a poor energy source, with the net energy accounting for only about 50 % of the gross energy (JUST 1982).

The diets in our experiment were designed on a total lysine basis. However, there is a marked difference between the reported digestibilities of synthetic lysine (LEIBHOLZ et al. 1986) and the lysine in peas (BURACZEWSKA et al. 1989) and rapeseed meal (SAUER and THACKER 1986), which resulted in different amounts of digestible lysine in the experimental diets. Our performance results indicate that the difference in the utilization of synthetic and protein-bound amino acids is much smaller than expected, as also confirmed in the recent studies of JORGENSEN and FERNANDEZ (1988), SUSENBETH et al. (1991) and MATRE and HOMB (1991).

Our calculations concerning the reduction in nitrogen excretion showed differences of about 0.63, 1.24 and 1.67 kg (14.7, 28.4 and 38.3%) in nitrogen excretion per pig between the control group and treatments 2, 3 and 4, respectively (Table 3). The results are in agreement with the calculations of LENIS (1989), who found that a reduction of 2 percentage units in the crude protein content of diets of growing pigs resulted in a reduction in nitrogen excretion of about 25%. JONGBLOED and LENIS (1992) have also calculated that nitrogen output could be lowered by 1 kg per pig by restricting the protein level of the diet and by adding synthetic lysine and methionine.

Gilts and castrated males showed similar growth performance in our experiment, although castrated

males deposited more fat compared to gilts. A higher rate of fat deposition in castrated males is usually accompanied by reduced growth (YEN et al. 1986). However, results similar to ours have been obtained in earlier studies (TAYLOR et al. 1979, ALAVIUHKOLA et al. 1992). It is possible that the amount of lysine in our trial diets was not sufficient for the gilts to show their full performance.

In conclusion, the results show that it is possible

to reduce the crude protein content of the diets of growing-finishing pigs by up to 20% with the addition of synthetic lysine and threonine, resulting in a reduction of about 30% in nitrogen excretion. However, the practical implementation of the feeding strategy depends on the price difference between the protein and energy feedstuffs and on the availability of synthetic amino acids at competitive prices.

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## SELOSTUS

### Lihaskojen rehun raakavalkuaisen täydentäminen puhtailla aminohapoilla

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Maatalouden tutkimuskeskus

Lihaskojen kasvatuskokeessa tutkittiin dieetin raakavalkuaispitoisuuden alentamisen ja synteettisen lysiini- ja treoniinitäydennyksen vaikutuksia tuotantotuloksiin ja teuraslaatuun. Yhteensä 120 sikaa, joista puolet imisiä ja puolet leikkoja, jaettiin neljään yhtä suureen ryhmään. Siat pidettiin kolmen eläimenkarsinoissa, joissa sukupuolet olivat erillään. Rajoitettu iänmukainen ruokintanormi oli 1,2-3,0 ry/sika/pv. Sikojen keskimääräinen alkupaino oli 25,5 kg ja loppupaino teuraaksi lähetettäessä 105,0 kg.

Vertailuryhmän rehuseos koostui ohrasta, rypsirouheesta, herneestä sekä kivennäis- ja vitamiiniseoksesta ja sisälsi raakavalkuaista 179 g/ry (sulavaa raakavalkuaista (SRV) 141 g/ry). Kolmen koeryhmän dieeteissä vähennettiin rypsirouheen ja herneen osuutta lineaarisesti, mutta rehuseosten lysiini- ja treoniinitasot pidettiin samana synteettisen lysiini- ja treoniinilisäyksen avulla (lysiiniä 9,0 g/ry ja treoniinia 5,9-6,4 g/ry). Dieettien raakavalkuaispitoisuus oli 160, 140 ja 122 g/ry (SRV 126, 111 ja 97 g/ry).

Raakavalkuaispitoisuuden alentamisella ei ollut vaikutusta sikojen päiväkasvuun tai rehuhyötysuhteeseen, kun dieetin lysiini- ja treoniinimäärät pidettiin samana. Valkuaispitoisuuden alentaminen heikensi kuitenkin suoraviivaisesti sikojen teuraslaatua. Selkä- ja kylkilavan paksuus kasvoi ja lihan osuus koko ruhossa sekä sen arvokkaissa osissa väheni. Käytännössä erot olivat kuitenkin varsin pieniä valkuaiastason 140 g/ry asti. Leikkojen tai imisien välillä ei havaittu eroja päiväkasvussa tai rehuhyötysuhteessa, mutta leikkojen teuraslaatu oli selvästi heikompi kuin imisien.

Kokeen tulosten perusteella rehun raakavalkuaispitoisuutta voidaan alentaa ainakin 20 %, kun dieetin aminohappokoostumusta samalla täydennetään sopivasti synteettisen lysiinin ja treoniinin avulla. Samalla vähennetään myös virtsan ja sonnan mukana vapautuvan typen määrää. Menetelmän käyttö on kuitenkin käytännössä riippuvainen eri rehukomponenttien hintasuhteista.