

# Optimum proportion of standardized ileal digestible sulfur amino acid to lysine to maximize the performance of 25–50 kg growing pigs fed reduced crude protein diets fortified with amino acids

G.J. ZHANG<sup>1</sup>, P.A. THACKER<sup>2</sup>, J.K. HTOO<sup>3</sup>, S.Y. QIAO<sup>1</sup>

<sup>1</sup>State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, P.R. China

<sup>2</sup>Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Canada

<sup>3</sup>Evonik Industries AG, Nutrition Research, Hanau-Wolfgang, Germany

**ABSTRACT:** The study was conducted to determine the standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) ratio required to maximize the performance of 25–50 kg pigs fed reduced crude protein (CP) diets fortified with crystalline amino acids. A total of 360 crossbred (Duroc × (Landrace × Large White)) pigs, weighing  $25.6 \pm 2.7$  kg, were blocked by gender, litter, and initial body weight (BW) and allotted to 1 of 5 dietary treatments with 6 pens per treatment and 12 pigs per pen for a 35-day performance trial. The basal diet was based on corn, soybean meal, and wheat bran and was formulated to be deficient in SAA (50% proportion of SID SAA to Lys). Graded levels of DL-methionine were added to the basal diet at the expense of wheat bran in order to provide 55.6, 60.0, 65.6, or 70.0% proportion of SID SAA to Lys, respectively. A constant SID Lys level of 0.90% was set so that Lys was the second limiting amino acid (AA) in all diets. Average daily gain (ADG) and feed conversion ratio (FCR) improved (linear and quadratic,  $P < 0.05$ ) with increasing dietary proportion of SID SAA to Lys. Increasing the dietary proportion of SID SAA to Lys decreased the serum urea nitrogen (SUN) level (quadratic,  $P < 0.05$ ). A two-slope broken-line model estimated the optimum proportion of SID SAA to Lys to be 62.2, 61.5, and 62.3% for maximum ADG and minimum FCR and SUN, respectively, whereas a curvilinear-plateau model yielded an optimum proportion of SID SAA to Lys level of 63.8, 62.5, and 61.5% for maximum ADG and minimum FCR and SUN, respectively. Based on an average of these estimates, we conclude that the proportion of SID SAA to Lys required for 25–50 kg pigs fed low CP diets is 62.3%. This estimate is higher than the NRC (2012) recommendation of 56.1% for 25–50 kg pigs fed normal CP diets.

**Keywords:** amino acid ratio; requirement

**List of abbreviations:** SID = standardized ileal digestible, SAA = sulfur amino acid, Lys = lysine, Met = methionine, Cys = cysteine, Trp = tryptophan, Val = valine, CP = crude protein, BW = body weight, AA = amino acid, ADG = average daily gain, FCR = feed conversion ratio, ADFI = average daily feed intake, SUN = serum urea nitrogen, NE = net energy

## INTRODUCTION

Methionine (Met) is generally considered to be the second or third limiting amino acid (AA) in typical corn-soybean meal based diets fed to grow-

ing pigs (Gaines et al. 2005; Yi et al. 2006). Met is an essential AA for normal growth which cannot be synthesized in the body, whereas cysteine (Cys) can be converted from Met as needed (Lu et al. 2009). Hence, the amount of Met needed in the

Supported by the National Key Basic Research Program of China Plan of P.R. China (2012CB124702).

doi: 10.17221/8276-CJAS

diet depends on the amount of Cys present. Thus, it is important to have requirement estimates for Met plus Cys or the sulfur AA (SAA) in addition to Met when formulating swine diets.

Lowering the crude protein (CP) content and supplementing the diet with crystalline AA has nutritional, economic, and environmental benefits (Kerr et al. 2003; Chen et al. 2011). However, for the successful application of reduced CP diets, it is critical to have reliable estimates of the optimum proportion of SAA to lysine (Lys) for pigs.

Previously reported optimum proportions of standardized ileal digestible (SID) SAA to Lys for 28–49 kg growing pigs range from 57.3 to 63.5% depending on the response parameters used (Yi et al. 2005), and are slightly higher than the NRC (2012) recommendation of 56.0% for 25–50 kg pigs. However, data estimating the dietary proportion of SID SAA to Lys ratio for 25–50 kg pigs fed reduced CP diets fortified with AA are scarce. Therefore, the present experiment was conducted to determine the proportion of SID SAA to Lys ratio required to maximize the performance of 25–50 kg pigs fed reduced CP diets fortified with AA. We hypothesized that the optimum proportion of SID SAA to Lys ratio for growing pigs receiving low CP diets differs from that of pigs fed normal CP diets.

## MATERIAL AND METHODS

### *Animals, housing, and dietary treatments.*

The China Agricultural University Animal Care and Use Committee (Beijing, China) reviewed and approved all animal protocols used in the present research.

A total of 360 crossbred (Duroc × (Landrace × Large White)) pigs, with an average initial body weight (BW) of  $25.6 \pm 2.7$  kg (mean ± SD), were used in the present study. The pigs were housed in a commercial growing facility located at the National Feed Engineering Technology Research Center (Beijing, China). Pigs were transferred from the nursery room to the growing facility, blocked by gender, litter, and initial BW and then allotted to 1 of 5 dietary treatments with 6 pens per treatment (3 pens of barrows and 3 pens of gilts) and 12 pigs per pen. The  $4.2 \times 4.6$  m pens were equipped with nipple drinkers and a four-hole self-feeder and had a totally slatted, concrete floor.

The pigs received a commercial starter diet for the first 7 days following transfer and the commercial

diet was progressively replaced by the experimental diets during the next 3 days. The experiment started on day 11 after transfer to the growing facility. The basal diet was based on corn, soybean meal, wheat bran, and crystalline AA (Lys, Met, threonine, tryptophan, isoleucine, and valine) and was formulated to be deficient in SAA (50% proportion of SID SAA to Lys). Graded levels of DL-Met were added to the basal diet at the expense of wheat bran to provide 55.6, 60.0, 65.6 or 70.0% proportions of SID SAA to Lys, respectively.

A constant SID Lys level of 0.90% was provided so that Lys was the second limiting AA in all diets in order to avoid an underestimation of the proportion of SID SAA to Lys. A SID Lys content of 0.90% is approximately 90% of the requirement determined in previous experiments for pigs of similar genetics and BW range (Zhang et al. 2012, 2013). The remaining AAs (except Lys and SAA) were supplemented to provide 105% of their requirement relative to Lys (NRC 2012). All diets contained 2.4 Mcal/kg of net energy (NE) and were fed in mash form. Pigs were fed *ad libitum* and allowed free access to water. Pigs and feeders were weighed at trial initiation and termination (day 35) to determine average daily gain (ADG), average daily feed intake (ADFI), and feed conversion ratio (FCR).

At the end of the experiment (day 35), blood samples were collected from the anterior *vena cava* of 2 pigs per pen into 10-ml heparin-free vacutainer tubes (Becton Dickinson Vacutainer Systems, Franklin Lakes, USA) after an overnight fast (12 h). The blood samples were centrifuged (3000 g at 4°C for 10 min) and serum was collected and frozen (–80°C) until an analysis of serum AA and urea nitrogen (SUN) could be completed.

**Chemical analyses.** The CP (AOAC method 984.13), ether extract (AOAC method 920.39), crude fibre (AOAC method 978.19), calcium (AOAC method 968.08), and phosphorus (AOAC method 965.17) contents of the ingredients and diets were analyzed according to the procedures of the Association of Official Analytical Chemists (AOAC 2003). For the analysis of most AA, ingredients and diets were hydrolyzed with 6 N HCl at 110°C for 24 h (AOAC 2003). For analysis of the sulfur-containing AA, performic acid oxidation preceded acid hydrolysis whereas the tryptophan (Trp) content was determined after alkaline hydrolysis (AOAC 2003). The AA analyses were performed using High Performance Liquid

Table 1. Ingredient composition of the experimental diets used to determine the optimal proportion of standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) for 25–50 kg pigs (as-fed basis)

Ingredients (g/kg)	Proportion of SID SAA to Lys (%)				
	50.0	55.6	60.0	65.6	70.0
Corn	744.2	744.2	744.2	744.2	744.2
Soybean meal (430 g/kg CP)	120.0	120.0	120.0	120.0	120.0
Wheat bran	90.0	89.5	89.0	88.5	88.0
Soybean oil	5.0	5.0	5.0	5.0	5.0
Limestone	11.0	11.0	11.0	11.0	11.0
Dicalcium phosphate	6.3	6.3	6.3	6.3	6.3
Salt	4.0	4.0	4.0	4.0	4.0
Vitamin-mineral premix <sup>1</sup>	10.0	10.0	10.0	10.0	10.0
L-Lysine HCl	4.9	4.9	4.9	4.9	4.9
DL-Methionine	0.6	1.1	1.6	2.1	2.6
L-Threonine	1.8	1.8	1.8	1.8	1.8
L-Tryptophan	0.4	0.4	0.4	0.4	0.4
L-Isoleucine	0.7	0.7	0.7	0.7	0.7
L-Valine	1.1	1.1	1.1	1.1	1.1

<sup>1</sup>premix provided the following per kg of complete diet for growing pigs: vitamin A 5.512 IU, vitamin D<sub>3</sub> 2200 IU, vitamin E 64 IU, vitamin K<sub>3</sub> 2.2 mg, vitamin B<sub>12</sub> 27.6 µg, riboflavin 5.5 mg, pantothenic acid 13.8 mg, niacin 30.3 mg, choline chloride 551 mg, Mn 40 mg, Fe 100 mg, Zn 100 mg, Cu 100 mg, I 0.3 mg, Se 0.3 mg

Chromatography (HPLC) (L-8800 AA Analyzer; Hitachi, Tokyo, Japan).

The AA content of the serum samples was determined by HPLC (L-8800 AA Analyzer; Hitachi) using pre-column derivatization with *o*-phthalaldehyde (Jones and Gilligan 1983). All chromatographic procedures were performed at room temperature and the samples and standards were evaluated in duplicate as described by Sedgwick et al. (1991). SUN concentration was measured using a Biochemical Analytical Instrument (Bayer Diagnostics Manufacturing Ltd., Dublin, Ireland) according to the method of Tao et al. (1982).

**Statistical analyses.** Data were analyzed by ANOVA using the GLM and PROC NLIN procedures of SAS (Statistical Analysis System, Version 9.1, 2002). Sex of pig was removed from the statistical model because there were no sex effects. Each pen was considered as a separate experimental unit. Orthogonal polynomial contrasts were used to determine linear and quadratic effects of increasing dietary proportion of SID SAA to Lys when the ANOVA indicated that significant differences were present. An alpha level of  $P < 0.05$  was set as the criterion for statistical significance. The optimal proportion of SID SAA to Lys was estimated by

subjecting the treatment Least Squares Means for ADG, FCR, and SUN to a two-slope, broken-line and a curvilinear-plateau regression model, based on best fit, as described by Robbins et al. (2006).

## RESULTS

No signs of illness, abnormal behaviour or mortality were observed during the current study. The effects of proportion of SID SAA to Lys on pig performance are presented in Table 3. The ADG and FCR were improved (linear and quadratic,  $P < 0.05$ ) as the dietary proportion of SID SAA to Lys increased from 50.0 to 70.0%. The ADFI was not different among the treatments. The best ADG and FCR were achieved at a proportion of SID SAA to Lys of 65.6%.

The effects of proportion of SID SAA to Lys on serum concentrations of AA and SUN are shown in Table 4. Dietary SAA supply significantly altered the serum concentrations of Lys, Met, Cys, and SUN. With increasing proportion of SID SAA to Lys, serum levels of Lys decreased linearly and quadratically ( $P < 0.05$ ), and the serum concentrations of Met and Cys increased linearly ( $P < 0.05$ ). The SUN concentration decreased as dietary proportion

doi: 10.17221/8276-CJAS

Table 2. Chemical analysis and calculated nutritional content of the experimental diets used to determine the optimal proportion of standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) for growing (25–50 kg) pigs (as-fed basis)<sup>1</sup>

	Proportion of SID SAA to Lys (%)				
	50.0	55.6	60.0	65.6	70.0
<b>Chemically determined values</b> (g/kg)					
Crude protein	140.2	140.6	141.0	141.4	141.7
Crude fibre	35.5	35.3	35.2	35.3	35.0
Ether extract	26.8	26.8	26.7	26.7	26.6
Calcium	6.0	6.0	6.0	6.0	6.0
Total phosphorus	4.6	4.6	4.6	4.6	4.6
Arginine	7.9	7.9	7.9	7.9	7.9
Histidine	3.5	3.5	3.5	3.5	3.5
Isoleucine	5.8	5.8	5.8	5.8	5.8
Leucine	12.4	12.4	12.4	12.4	12.4
Lysine	9.8	9.8	9.8	9.8	9.8
Methionine	2.7	3.2	3.7	4.1	4.6
Cystine	2.4	2.4	2.4	2.4	2.4
Threonine	6.6	6.6	6.6	6.6	6.6
Tryptophan	1.9	1.9	1.9	1.9	1.9
Phenylalanine	6.2	6.2	6.2	6.2	6.2
Valine	7.3	7.3	7.3	7.3	7.3
<b>Calculated values</b>					
NE (Mcal/kg) <sup>2</sup>	2.4	2.4	2.4	2.4	2.4
<b>SID amino acids</b> (g/kg) <sup>3</sup>					
Lysine	9.0	9.0	9.0	9.0	9.0
Methionine	2.4	2.9	3.3	3.8	4.2
Cystine	2.1	2.1	2.1	2.1	2.1
Threonine	5.8	5.8	5.8	5.8	5.8
Tryptophan	1.7	1.7	1.7	1.7	1.7
Leucine	10.6	10.6	10.6	10.6	10.6
Isoleucine	5.0	5.0	5.0	5.0	5.0
Valine	6.2	6.2	6.2	6.2	6.2
Histidine	3.1	3.1	3.1	3.1	3.1
Phenylalanine	5.3	5.3	5.3	5.3	5.3

<sup>1</sup>values are adjusted to a dry matter content of 87.5%<sup>2</sup>net energy (NE) values based on Noblet et al. (1994) where  $NE = (0.703 \times \text{kcal/kg digestible energy}) + (1.58 \times \text{ether extract}) + (0.47 \times \text{starch}) - (0.97 \times \text{crude protein}) - (0.98 \times \text{crude fibre})$  (nutrients in g/kg DM)<sup>3</sup>values for SID concentrations of amino acids (AA) for the ingredients were obtained from Sauvant et al. (2004) and AminoDat (Version 3.0, 2005). SID levels in the diets were calculated by multiplying the SID AA content of the individual ingredients by their inclusion level in the diets and then summing the products

of SID SAA to Lys increased (linear and quadratic,  $P < 0.05$ ) and the SUN concentration was the lowest at a proportion of SID SAA to Lys of 0.656 (Table 4).

A two-slope broken-line regression analysis estimated the optimum proportion of SID SAA to Lys to be 62.2, 61.5, and 62.3% to maximize ADG and minimize FCR and SUN, respectively. A curvilinear-plateau analysis estimated the op-

imum proportion of SID SAA to Lys to be 63.8, 62.5, and 61.5% to maximize ADG and minimize FCR and SUN, respectively (Figures 1–3).

## DISCUSSION

The present experiment was conducted to determine the optimum proportion of SID SAA to

Table 3. Effect of dietary proportion of standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) on the performance of growing (25–50 kg) pigs

	Proportion of SID SAA to Lys (%)					SEM	P-value <sup>1</sup>		
	50.0	55.6	60.0	65.6	70.0		ANOVA	linear	quadratic
Initial BW (kg)	25.9	25.8	25.7	25.7	25.6	1.23	0.92	ns	ns
Final BW (kg)	50.3	51.5	52.0	52.4	52.0	1.86	0.61	ns	ns
ADG (g/day)	697 <sup>a</sup>	738 <sup>b</sup>	751 <sup>bc</sup>	764 <sup>c</sup>	754 <sup>bc</sup>	7.9	0.03	0.04	0.02
ADFI (g/day)	1662	1667	1645	1650	1645	13.3	0.27	ns	ns
FCR	2.38 <sup>c</sup>	2.26 <sup>b</sup>	2.19 <sup>a</sup>	2.16 <sup>a</sup>	2.18 <sup>a</sup>	0.021	0.01	0.03	0.04

BW = body weight, ADG = average daily gain, ADFI = average daily feed intake, FCR = feed conversion ratio, SEM = standard error of the mean, ns = non-significant

<sup>1</sup>data are means of six pens per treatment; P-values indicate the effects from all treatments; linear and quadratic contrasts for proportion of SID SAA to Lys

<sup>a-c</sup>means in the rows with different letters are different ( $P < 0.05$ )

Lys for 25–50 kg growing pigs fed reduced CP diets fortified with crystalline AA. A constant 0.90% SID Lys level was maintained in all diets

used in the present study to ensure that the diets were marginally deficient in Lys as recommended for AA ratio dose-response trials (Boisen 2003).

Table 4. Effect of dietary proportion of standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) on the serum urea nitrogen and amino acid concentrations of growing (25–50 kg) pigs

	Proportion of SID SAA to Lys (%)					SEM <sup>1</sup>	P-value <sup>1</sup>	
	50.0	55.6	60.0	65.6	70.0		ANOVA	linear
Serum urea nitrogen (mmol/l)	4.81 <sup>a</sup>	3.73 <sup>bc</sup>	3.37 <sup>c</sup>	3.09 <sup>c</sup>	3.41 <sup>c</sup>	0.231	0.03	0.06
<b>Serum essential amino acids (nmol/ml)</b>								
Arginine	498	485	476	469	504	22.3	0.76	ns
Histidine	95	78	85	65	81	8.4	0.32	ns
Isoleucine	118	113	104	109	126	5.7	0.19	ns
Leucine	266	238	214	217	237	18.3	0.35	ns
Lysine	328 <sup>a</sup>	274 <sup>ab</sup>	261 <sup>ab</sup>	238 <sup>b</sup>	257 <sup>ab</sup>	13.5	0.04	0.03
Methionine	92 <sup>b</sup>	96 <sup>b</sup>	106 <sup>a</sup>	113 <sup>a</sup>	102 <sup>ab</sup>	3.8	0.02	0.04
Phenylalanine	111	95	90	105	114	6.7	0.46	ns
Threonine	271	251	226	203	227	20.6	0.12	ns
Tryptophan	72	60	51	47	59	6.4	0.33	ns
Valine	321	280	274	287	306	24.4	0.72	ns
<b>Serum non-essential amino acids (nmol/ml)</b>								
Alanine	503	460	441	428	457	32.8	0.21	ns
Aspartate	298	280	273	234	306	18.9	0.10	ns
Cysteine	67 <sup>b</sup>	76 <sup>ab</sup>	81 <sup>ab</sup>	93 <sup>a</sup>	97 <sup>a</sup>	7.3	0.04	0.03
Glutamate	296	259	236	237	309	26.8	0.21	ns
Glutamine	452	407	382	383	423	18.6	0.25	ns
Glycine	1322	1292	1235	1201	1258	61.8	0.69	ns
Serine	254	233	228	218	269	15.3	0.31	ns
Proline	436	394	370	417	453	25.7	0.57	ns

SEM = standard error of the mean, ns = non-significant

<sup>1</sup>data are means of six replicates per treatment; P-values indicate the effects from all treatments; linear and quadratic contrasts for proportion of SID SAA to Lys

<sup>a-c</sup>means in the rows with different letters are different ( $P < 0.05$ )

doi: 10.17221/8276-CJAS

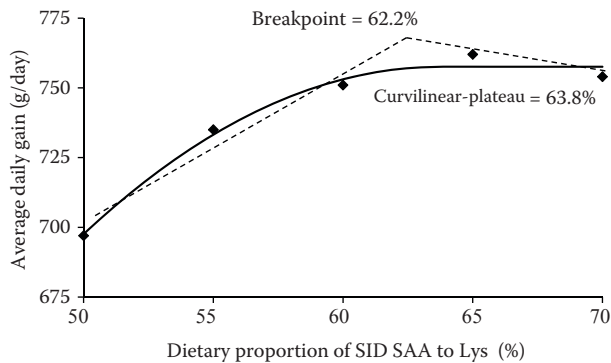


Figure 1. Fitted two-slope broken-line (---) and curvilinear-plateau (—) plot of average daily gain as a function of proportion of standardized ileal digestible (SID) sulfur amino acid (SAA) to lysine (Lys) with observed treatment mean values. Data points (♦) represent treatment means ( $n = 6$  observations per treatment mean). The minimal proportion of SID SAA to Lys estimated by a two-slope broken-line was 62.2% ( $y = 766.5 - 5.4(62.2 - x) - 1.6(x - 62.2)$ ;  $R^2 = 0.97$ ). The curvilinear-plateau model estimated the optimal proportion of SID SAA to Lys to be 63.8% ( $y = 757.6 - 0.32(63.8 - x)^2$ ;  $R^2 = 0.99$ ).

In the current study, ADFI was not affected by treatment but ADG and FCR improved linearly and quadratically with increasing proportion of SID SAA to Lys and were optimized at a proportion of SID SAA to Lys of 65.6%. These results agree with Yi et al. (2005) who reported linear and quadratic effects of an increasing proportion of SID SAA to Lys on ADG and gain : feed which optimized at 64.5% proportion of SID SAA to Lys for 28–49 kg pigs.

Changes in the plasma concentration of AA resulting from increasing the dietary supply of a test AA can be considered as indicative of the optimum AA supply to pigs (Wiltafsky et al. 2009). In the current study, increasing dietary SAA supply decreased serum levels of Lys but linearly increased serum levels of Cys and Met, which was likely due to the fact that Met can be converted to Cys through the trans-sulfuration pathway but not *vice versa* (Lu 2009). Wiltafsky et al. (2009) suggested that increasing the dietary supply of valine (Val) decreased plasma Lys concentration but linearly increased plasma Val concentration.

The estimation of the optimum proportion of AA to Lys is affected by the statistical models used. There are different views on which of these models provides the best statistical fit (Robbins et al. 1979; Coma et al. 1995). Pomar et al. (2003) suggested that a linear broken-line model should be used

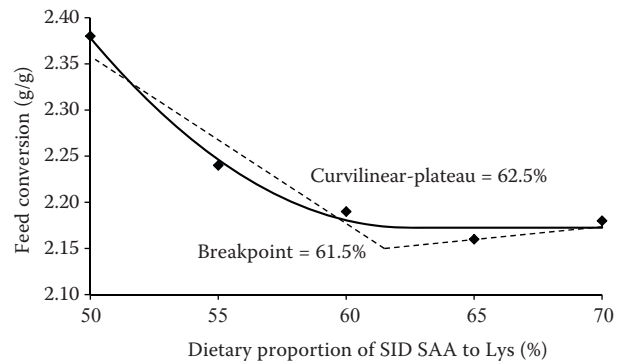


Figure 2. Fitted two-slope broken-line (---) and curvilinear-plateau (—) plot of feed conversion ratio as a function of proportion of SID sulfur amino acid (SAA) to lysine (Lys) with observed treatment mean values.

Data points (♦) represent treatment means ( $n = 6$  observations per treatment mean). The minimal proportion of SID SAA to Lys estimated by a two-slope broken-line was 61.5% ( $y = 2.15 + 0.019(61.5 - x) + 0.004(x - 61.5)$ ;  $R^2 = 0.96$ ). The curvilinear-plateau model estimated the optimal proportion of SID SAA to Lys to be 62.5% ( $y = 2.17 + 0.001(62.5 - x)^2$ ;  $R^2 = 0.99$ ).

for individual animals, while a curvilinear-plateau model is best suited for a population or group of animals. The quadratic broken-line (curvilinear plateau) model or a combination of quadratic and broken-line models has been used to estimate AA

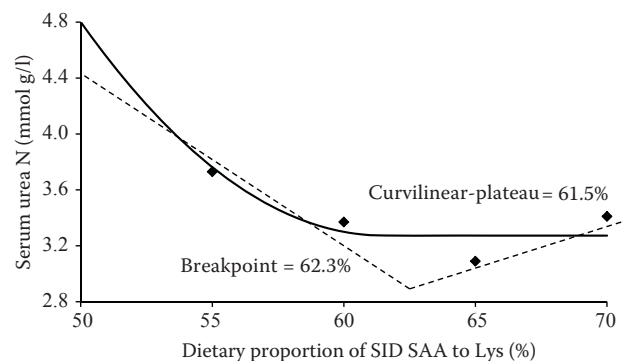


Figure 3. Fitted two-slope broken-line (linear-breakpoint) (---) and curvilinear-plateau (—) plot of serum urea nitrogen concentration as a function of proportion of SID sulfur amino acid (SAA) to lysine (Lys) with observed treatment mean values.

Data points (♦) represent treatment means ( $n = 6$  observations per treatment mean). The minimal proportion of SID SAA to Lys estimated by a two-slope broken-line was 62.3% ( $y = 2.92 + 0.144(62.3 - x) + 0.064(x - 62.3)$ ;  $R^2 = 0.95$ ). The curvilinear-plateau model estimated the optimal proportion of SID SAA to Lys to be 61.5% ( $y = 3.27 + 0.012(61.5 - x)^2$ ;  $R^2 = 0.97$ ).

requirements for curvilinear data sets (Robbins et al. 2006). In the present study, we used the average of the estimates derived from the two-slope broken-line and curvilinear-plateau models to minimize the variation obtained from the statistical models. Interestingly, the estimates from these two models were similar. The two-slope broken-line model estimated the optimum proportion of SID SAA to Lys to be 62.2, 61.5, and 62.3% to maximize ADG and minimize FCR and SUN, respectively, whereas the curvilinear-plateau model yielded an optimum proportion of SID SAA to Lys of 63.8, 62.5, and 61.5% to maximize ADG and minimize FCR and SUN, respectively. Averaging these estimates, an optimal proportion of SID SAA to Lys of 62.3% was obtained which is higher than the NRC (2012) recommendation of 56.1% for 25–50 kg pigs but agrees with Yi et al. (2005), based on a broken-line model using the intercept of the broken-line and quadratic curve, and the 95% upper asymptote of quadratic response. They estimated the optimum proportion of SID SAA to Lys to be 61.7% for ADG and 61.1% for gain : feed in 28–49 kg mixed-sex pigs. Similarly, Gaines et al. (2004) estimated the optimum proportion of SID SAA to Lys to be 59.7% and 61.1% for ADG and gain : feed ratio for 29–45 kg growing pigs. Wang and Fuller (1989), using the deletion method and N-retention as the response criteria, estimated the proportion of ideal digestible SAA to Lys for 25–50 kg pigs to be 63.0%. In a literature review, Peak (2005) reported the proportion of SID SAA to Lys to be 60.0–62.0% for modern genotypes of nursery and growing pigs.

The optimal proportion of SAA to Lys can be affected by several factors. Firstly, the ratio of Met to Cys in the diet can affect the proportion of SAA to Lys because Cys can be converted from Met but not *vice versa*, i.e. the level of Cys can affect the total SAA requirement. Chung and Baker (1992) reported that Cys can furnish up to 50% of the young pig's SAA requirement. Shoveller et al. (2003) found that when Met is meeting 100% of the SAA requirement, Cys can replace 40% of the Met requirement. More recently, Gillis et al. (2007) and Qiao et al. (2008) suggested an optimal proportion of Met to SAA of 55.0% for growing pigs. In the basal diet of the current study, the SID Met : Cys ratio was 53 : 47 which is similar to the ratio that Yi et al. (2006) used to determine the SID SAA requirement for nursery pigs.

Secondly, SAA are also needed for synthesis of compounds involved in the immune response

such as glutathione and acute phase proteins such as albumin and fibrinogen which are high in Cys (Reeds et al. 1994; Malmezat et al. 2000; Kim et al. 2012; Litvak et al. 2013). Recent research showed that growing pigs, exposed to an immune challenge or poor health status, have an increased requirement for SAA (Rakhshandeh et al. 2010; Kim et al. 2012). The current experiment was conducted in a commercial facility and no in-feed antibiotics were included in the diet. Thus, it represents a low sanitary or immune status which might have influenced the derived proportion of SAA to Lys.

The optimum proportion of SAA to Lys estimated from the performance data in the current experiment were in line with the proportion of SAA to Lys derived from the SUN data. In the current experiment, the lowest SUN concentration was observed at a proportion of SAA to Lys of 65.6%. Urea is the main end product originating from the catabolism of AA and a reduced SUN is indicative of a reduced need for deamination of excess AA (D'Mello 2003). The concentration of SUN has been used as response criteria for determination of AA requirements (Pedersen and Boisen 2001).

Using the overall results of this experiment, the two-slope broken-line model estimated the optimum proportion of SAA to Lys to be 62.2, 61.5, and 62.3% for ADG, FCR, and SUN, respectively, whereas the curvilinear-plateau model yielded an optimum proportion of SAA to Lys of 63.8, 62.5, and 61.5% based on ADG, FCR, and SUN, respectively. Based on an average of these estimates, we conclude that the optimal proportion of SAA to Lys for pigs fed low CP diets supplemented with crystalline AA is 62.3% for 25–50 kg pigs. This estimate is higher than the NRC (2012) recommendation of 56.1% for 25–50 kg pigs fed normal CP diets. We suggest that crystalline AA may be more susceptible to reactions with other compounds in the feed than protein-bound AA thus resulting in a higher requirement for SAA when low CP diets supplemented with crystalline AA are fed. In particular, the availability of free Met may be reduced by reactions with radicals produced from the oxidation of unsaturated fatty acids in the feed (Boisen 2003).

## CONCLUSION

In conclusion, the proportion of SAA to Lys required to maximize ADG and minimize FCR of 25–50 kg pigs receiving low CP diets supplemented

doi: 10.17221/8276-CJAS

with crystalline AA was calculated to be 62.3%. The serum urea N data agree with the estimated optimal proportion of SAA to Lys established for maximum pig performance. It appears that the proportion of SAA to Lys required for pigs fed low CP diets supplemented with crystalline AA may be higher than that required for pigs fed normal CP diets.

**Acknowledgement.** The authors thank Feed Additives, Evonik Industries AG for providing supplemental amino acids.

## REFERENCES

- AOAC (2003): Official Methods of Analysis. 17<sup>th</sup> Ed. Association of Official Analytical Chemists, Arlington, USA.
- Boisen S. (2003): Ideal dietary amino acid profiles for pigs. In: D'Mello J.P.F. (ed.): Amino Acids in Animal Nutrition. CABI Publishing, Edinburgh, UK, 157–186.
- Chen H.Y., Yi X.W., Zhang G.J., Lu N., Chu L.C., Thacker P.A., Qiao S.Y. (2011): Studies on reducing nitrogen excretion: 1. Net energy requirements of finishing pigs maximizing performance and carcass quality fed low crude protein diets supplemented with crystalline amino acids. *Journal of Animal Science and Biotechnology*, 2, 84–93.
- Chung T.K., Baker D.H. (1992): Ideal amino acid pattern for 10-kilogram pigs. *Journal of Animal Science*, 70, 3102–3111.
- Coma J., Carrion D., Zimmerman D.R. (1995): Use of plasma urea nitrogen as a rapid response criterion to determine the lysine requirement of pigs. *Journal of Animal Science*, 73, 472–476.
- D'Mello J.P.F. (2003): An outline of pathways in amino acid metabolism. In: D'Mello J.P.F. (ed.): Amino Acids in Animal Nutrition. CABI Publishing, Edinburgh, UK, 71–86.
- Gaines A.M., Yi G.F., Ratliff B.W., Srichana P., Alle G.L., Knight C.D., Perryman K.P. (2004): Estimation of the true ileal digestible sulfur amino acid : lysine ratio for growing pigs weighing 29–45 kilograms. *Journal of Animal Science*, 83 (Suppl. 1), 294.
- Gaines A.M., Yi G.F., Ratliff B.W., Srichana P., Kendall D.C., Allee G.L., Knight C.D., Perryman K.R. (2005): Estimation of the ideal ratio of true ileal digestible sulfur amino acids : lysine in 8- to 26-kg nursery pigs. *Journal of Animal Science*, 83, 2527–2534.
- Gillis A.M., Reijmers A., Pluske J.R., de Lange C.F.M. (2007): Influence of dietary methionine to methionine plus cysteine ratios on nitrogen retention in gilts fed purified diets between 40 and 80 kg live body weight. *Canadian Journal of Animal Science*, 87, 87–92.
- Jones B.N., Gilligan J.P. (1983): o-Phthalaldehyde precolumn derivatization and reversed phase high performance liquid chromatography of polypeptide hydrolysates and physiological fluids. *Journal of Chromatography*, 266, 471–482.
- Kerr B.J., Southern L.L., Bidner T.D., Friesen K.G., Easter R.A. (2003): Influence of dietary protein level, amino acid supplementation, and dietary energy levels on growing-finishing pig performance and carcass composition. *Journal of Animal Science*, 81, 3075–3087.
- Kim J.C., Mullan B.P., Frey B., Payne H.G., Pluske J.R. (2012): Whole body protein deposition and plasma amino acid profiles in growing and/or finishing pigs fed increasing levels of sulfur amino acids with and without *Escherichia coli* lipopolysaccharide challenge. *Journal of Animal Science*, 90, 362–365.
- Litvak N., Rakhshandeh A., Htoo J.K., de Lange C.F.M. (2013): Immune system stimulation increases the optimal dietary methionine to methionine plus cysteine ratio in growing pigs. *Journal of Animal Science*, 91, 4188–4196.
- Lu S.C. (2009): Regulation of glutathione synthesis. *Molecular Aspects of Medicine*, 30, 42–59.
- Malmezat T., Breuille D., Capitan P., Mirand P.P., Obléd C. (2000): Glutathione turnover is increased during the acute phase of sepsis in rats. *Journal of Nutrition*, 130, 1239–1246.
- Noblet J., Fortune H., Shi X.S., Dubois S. (1994): Prediction of net energy value of feeds for growing pigs. *Journal of Animal Science*, 72, 344–354.
- NRC (2012): Nutrient Requirements of Swine. 11<sup>th</sup> Ed. The National Academies Press, Washington, USA.
- Peak S. (2005): TSAA requirement for nursery and growing pigs. In: Foxcroft G. (ed.): Advances in Pork Production. University of Alberta Press, Edmonton, Canada, 101–107.
- Pedersen C., Boisen S. (2001): Studies on the response time for plasma urea nitrogen as a rapid measure for dietary protein quality in pigs. *Animal Science*, 51, 209–216.
- Pomar C., Kyriazakis I., Emmans G.C., Knap P.W. (2003): Modeling stochasticity: dealing with populations rather than individual pigs. *Journal of Animal Science*, 81, E178–E186.
- Qiao S., Piao X., Feng Z., Ding Y., Yue L., Thacker P.A. (2008): The optimum methionine to methionine plus cysteine ratio for growing pigs determined using plasma urea nitrogen and nitrogen balance. *Asian-Australasian Journal of Animal Sciences*, 21, 434–442.
- Rakhshandeh A., Htoo J.K., de Lange C.F.M. (2010): Immune system stimulation of growing pigs does not alter apparent ileal amino acid digestibility but reduces the ratio between whole body nitrogen and sulfur retention. *Livestock Science*, 134, 21–23.



- Reeds P.J., Fjeld C.R., Jahoor F. (1994): Do the differences between the amino acid compositions of acute-phase and muscle proteins have a bearing on nitrogen loss in traumatic states? *Journal of Nutrition*, 124, 906–910.
- Robbins K.R., Norton H.W., Baker D.H. (1979): Estimation of nutrient requirements from growth data. *Journal of Nutrition*, 109, 1710–1714.
- Robbins K.R., Saxton A.M., Southern L.L. (2006): Estimation of nutrient requirements using broken-line regression analysis. *Journal of Animal Science*, 84, E155–E165.
- Sauvant D., Perez J.M., Tran G. (eds) (2004): *Tables of Composition and Nutritional Value of Feed Materials: Pigs, Poultry, Cattle, Sheep, Goats, Rabbits, Horses and Fish*. INRA Editions, Paris, France. (in French)
- Sedgwick G.W., Fenton T.W., Thompson J.R. (1991): Effect of protein precipitating agents on the recovery of plasma free amino acids. *Canadian Journal of Animal Science*, 71, 953–957.
- Shoveller A.K., Brunton J.A., Pencharz P.B., Ball R.O. (2003): The methionine requirement is lower in neonatal piglets fed parentally than in those fed enterally. *Journal of Nutrition*, 133, 1390–1397.
- Tao Y., Ying C., Ni Z., Li J., Li L. (1982): *Clinical Biochemical Analysis*. Shanghai Scientific Technical Press, Shanghai, China.
- Wang T.C., Fuller M.F. (1989): The optimum dietary amino acid pattern for growing pigs. *British Journal of Nutrition*, 62, 77–89.
- Wiltafsky M.K., Schmidlein B., Roth F.X. (2009): Estimates of the optimum dietary ratio of standardized ileal digestible valine to lysine for eight to twenty-five kilograms of body weight pigs. *Journal of Animal Science*, 87, 2544–2553.
- Yi G.F., Gaines A.M., Ratliff B.W., Srichana P., Alle G.L., Knight C.D., Perryman K.P. (2005): Estimation of the true ileal digestible sulfur amino acid:lysine ratio for growing pigs weighing 28–49 kilograms. *Journal of Animal Science*, 83 (Suppl. 1), 213.
- Yi G.F., Gaines A.M., Ratliff B.W., Srichana P., Allee G.L., Perryman K.R., Knight C.D. (2006): Estimation of the true ileal digestible lysine and sulfur amino acid requirement and comparison of the bioefficacy of 2-hydroxy-4-(methylthio) butanoic acid and DL-methionine in eleven- to twenty-six-kilogram nursery pigs. *Journal of Animal Science*, 84, 1709–1721.
- Zhang G.J., Song Q.L., Xie C.Y., Chu L.C., Thacker P.A., Htoo J.K., Qiao S.Y. (2012): Estimation of the ideal standardized ileal digestible tryptophan to lysine ratio for growing pigs fed low crude protein diets supplemented with crystalline amino acids. *Livestock Science*, 149, 260–266.
- Zhang G.J., Xie C.Y., Thacker P.A., Htoo J.K., Qiao S.Y. (2013): Estimation of the ideal ratio of standardized ileal digestible threonine to lysine for growing pigs (22–50 kg) fed low crude protein diets supplemented with crystalline amino acids. *Animal Feed Science and Technology*, 180, 83–91.

Received: 2014–09–11

Accepted after corrections: 2014–12–02

---

*Corresponding Author*

Dr. Shiyan Qiao, China Agricultural University, College of Animal Science and Technology, State Key Laboratory of Animal Nutrition, Haidian district, Beijing, 100193, P.R. China  
Phone: +86 10 6273 1456, e-mail: qiaoshy@mafic.ac.cn

---