

Protected lysine in diets for 25–100 kg pigs

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Abstract

The digestible lysine requirements for fattening pigs have not been established clearly. The objective of this trial was to determine a level of protected lysine in pig diets that was adequate for growth performance, carcass characteristics, and plasma urea nitrogen concentration. Fifty crossbred pigs (29.55 ± 1.80 kg initial bodyweight) were used in a completely randomized design with a 3×2 factorial arrangement of treatments. Treatments were 0.90%, 1.00% and 1.10% lysine in the growing phase, followed by diets with 0.75%, 0.85% and 0.95% lysine in finishing stage I, and 0.83%, 0.93% and 1.03% lysine in finishing stage II. The lysine was provided in its conventional form and as protected lysine. For growing pigs, the highest average daily feed intake (ADFI) and final bodyweight were obtained with 1.1% and 1.0% lysine, respectively. The average daily gain, ADFI, final bodyweight, fat free lean gain and *longissimus* muscle area (LMA) were reduced with protected lysine. In finishing I stage, pigs fed 0.95% lysine had greater final bodyweight, LMA, and lean meat percentage than pigs fed with 1% unprotected lysine. For finishing II pigs, ADFI and final bodyweight were the greatest when 1.03% lysine was provided, regardless the type of lysine that was fed. The plasma urea nitrogen increased with the 1.03% lysine diet and was reduced with protected lysine. Results indicate that the digestible lysine requirement for the fattening pig diets might be 0.10% higher than in current recommendations. The use of protected lysine affected growth negatively during the growing stage.

Keywords: absorption, bioavailability, bioefficacy, growth performance, synthetic amino acids

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Introduction

The probable deficits of amino acids in feedstuffs, feed costs, and nitrogen excretion may be overcome by replacing a portion of the dietary crude protein with crystalline amino acids (Gloaguen *et al.*, 2014; Wang *et al.*, 2020) in the diet for pigs. For fattening pigs, lysine is the first limiting amino acid in diets formulated from conventional ingredients, which may lead to lysine deficiency or excess, reduce its availability for protein synthesis, and limit growth (NRC, 2012). Increasing the efficiency of lysine use for protein synthesis would lower the dietary requirement in swine, thus decreasing the need for supplemental lysine, and decrease production costs (Gatrell *et al.*, 2017). Reducing lysine catabolism has been shown to reduce the requirement for it (Cleveland *et al.*, 2008).

Levels of lysine that are higher than NRC (2012) recommendations improve productive performance (Figueroa *et al.*, 2012; Zhang *et al.*, 2013; Goncalves *et al.*, 2017; Soto *et al.*, 2019). This may imply that the digestible lysine requirement is higher than that suggested by NRC (2012), or that the dietary lysine is not used completely. Synthetic-free lysine is more sensitive to acidic conditions and more rapidly absorbed in the digestive tract than the protein-bound amino acid (Kondos & Adri, 1982; Sato *et al.*, 1984; Stein *et al.*, 2007). The pronounced, but transient postprandial increase of plasma amino acid and peptide concentrations might adversely affect metabolic efficiency and amino acid oxidation (Dangin *et al.*, 2001). In addition, rapid absorption of supplemented free amino acids in the small intestine of pigs relative to those derived from dietary protein could lead to an asynchrony in their availability to organs and tissues (Chen, 2017). Improved nutrient synchronization may increase nitrogen retention in growing pigs (Van den Borne *et al.*, 2007).

Therefore, it is necessary to find ways to increase lysine bio-efficacy and nutrient synchronization in pig diets.

The use of protected amino acids that can resist degradation in the stomach, allowing for absorption from the intestines, could help overcome these limitations (Piva et al., 2007) and has been reported in ruminant animals with resultant improvements in their productive performance (Lee et al., 2012; Zanton et al., 2014; Awawdeh, 2016). In pigs, replacing HCl-Lysine with protected lysine improves the bio-efficacy of this amino acid, without affecting growth and carcass characteristics negatively (Prandini et al., 2013). Incorporating protected methionine in pig diets also produces favourable changes in daily weight gain, feed intake, and backfat thickness (Figueroa-Velasco et al., 2020). Sun et al. (2020a) suggested supplemental levels of crystalline lysine and DL-methionine for broiler chickens could be reduced effectively by approximately 20% with an encapsulated form with resulting improvements in amino acid utilization efficiency, with no detrimental effects on production. Prandini et al. (2013) stated that microencapsulated lysine, in comparison with unprotected synthetic lysine, improved the efficiency with which pigs used crude protein and amino acids. This improvement is a result of slower rates of release and absorption microencapsulated lysine compared with unprotected synthetic lysine and lysine that is bound to dietary protein. Sun et al. (2020b) observed that feeding encapsulated lysine and methionine in laying hens may improve the post-absorptive amino acid balance and allow for reduced levels of supplemental amino acids.

Because increased lysine bio-efficacy can improve growth in pigs, it was hypothesized that the dietary addition of protected lysine would represent an alternative source of this amino acid compared with traditional synthetic lysine. Additionally, it was hypothesized that the dietary concentration of lysine could be changed when protected lysine is incorporated into the diet. Therefore, the aim of this research was to determine the optimum level of protected lysine in fattening pig diets based on their growth, carcass characteristics, and plasma urea nitrogen concentration.

Material and Methods

The experimental procedures were performed following the recommendations of the CIOMS (2012) and the Mexican law (SAGARPA, 1999) for the use of animals in experimentation. The experiment was conducted at the Swine Unit of the Experimental Farm, Colegio de Postgraduados, located in Montecillo, State of Mexico (98° 48' 27" W, 19° 48' 23" N). The climate is temperate, semi-arid, with an average annual temperature of 15.9 °C, infrequent frosts, an average annual rainfall of 686 mm and an altitude of 2,241 m above sea level (García, 2004).

The treatments consisted of three levels of digestible lysine from two sources (conventional and protected) in three stages between weaning and harvest. The lysine levels were 0.90%, 1.00%, and 1.10% in the growing stage, 0.75%, 0.85%, and 0.95% in finishing stage I, and 0.83%, 0.93%, and 1.03% in finishing stage II. These levels corresponded to a control treatment, a level below those recommended by the NRC (2012) and a level above. The diet formulations for the growing stage, finishing stage I and finishing stage II and their nutritional contents are provided in supplemental tables S1, S2 and S3.

Fifty crossbred terminal line (Duroc × Yorkshire × Landrace) barrows were used in a completely randomized design in a 3 × 2 factorial arrangement of treatments with three levels of digestible lysine provided in two forms. There were eight or nine replicates of each treatment. The average initial bodyweight was 29.55 ± 2.22 kg. The experiment was conducted in three stages: growing for 35 days, finishing I for 28 days, and finishing II for 29 days. The pigs were housed individually in 1.2 × 1.5 m pens with concrete and plastic slat floors that were equipped with a single feeder and a nipple drinker. Feed and water were provided ad libitum.

The digestible lysine levels were obtained by adding synthetic lysine or protected lysine to the control diet to increase the concentration of this amino acid. The protected lysine contained 50% encapsulated L-lysine HCl (Ajiipro™-L, Ajinomoto, Chicago, Illinois, USA). Basal diets were based on sorghum-soybean meal supplemented with crystalline amino acids (L-Lysine-HCl, DL-Methionine (Evonik Industries, Parsippany, NJ, USA), L-threonine (Jefo Nutrition Inc, Saint-Hyacinthe, Québec, Canada) and L-Tryptophan (CPB Aurum, Mexico City)) to meet or exceed the nutritional requirements for each stage of growth (NRC, 2012). In the diet for finishing stage II feed, 5 ppm of ractopamine (Paylean, Elanco, Mexico) was added and the requirements were adjusted following the recommendations of the NRC (2012) when this beta-agonist is fed.

Average daily gain (ADG), average daily feed intake (ADFI), and feed to gain ratio (FGR) were calculated from the recorded measures of bodyweight and feed intake. On the first and last days of the experiment, backfat thickness and *longissimus* muscle area (LMA) were measured using the Sonovet 600 real time ultrasound system with a 3.5 MHz transducer (Medison, Inc., Cypress, California, USA). These data, along with initial bodyweight and final bodyweight, were used to determine the fat-free lean gain (FFLG) and the lean meat percentage using the NPPC equations (Burson & Berg, 2001). A blood sample

collected on the last day of each experimental stage was used to determine plasma urea nitrogen (plasma urea nitrogen) concentration.

The crude protein contents of the diets were established with the macro Kjeldahl method (AOAC, 2005). Their calcium and phosphorus were ascertained with atomic absorption spectrophotometry (Karl *et al.*, 1979) with a Perkin Elmer 4000 model instrument (Series Lambda 2, Perkin Elmer Inc., Norwalk, CT, USA). Blood samples (5 mL) were collected on the last day of each experimental stage using vacutainer tubes without anticoagulant, and were immediately refrigerated at 4 °C. Blood samples were centrifuged (Sigma 2-16k, Germany) at 3500 rpm for 20 min to obtain the serum, which was stored in Eppendorf tubes and kept in a freezer (EUR251P7W Tappan, Electrolux Home Products North America, Augusta, GA, 30907 EUA) at -20 °C until the plasma urea nitrogen concentration (Chaney & Marbach, 1962) was established with a spectrophotometer (Vary Cary UV, Victoria, Australia).

Shapiro-Wilk and Levene's tests were used to check the normality and homogeneity of variance of all variables. The data were subjected to analysis of covariance variance using the GLM procedure of SAS (SAS Institute Inc., Cary, North Carolina, USA) with initial bodyweight being used as the covariate. Tukey's test ($P \leq 0.10$) was used to compare treatment means.

Results and Discussion

The highest ADFI ($P = 0.02$) was obtained with 1.10% lysine in the diet and reduced ($P = 0.07$) with the addition of protected lysine to the diets of growing pigs (Table 1). Average daily gain was reduced ($P = 0.10$) when the protected lysine was used and the level of protected lysine did not affect it ($P > 0.10$). Final bodyweight was also reduced by the addition of protected lysine ($P = 0.01$). The heaviest final bodyweight was observed ($P = 0.09$) with 1.00% of regular lysine. The FFLG and LMA were affected ($P \leq 0.05$) by protected lysine. The other variables were not influenced ($P > 0.10$) by the level or source of lysine. In the finishing I stage, pigs fed with 0.95% digestible lysine had heavier final bodyweight, larger LMA and higher lean meat percentage than those fed with the control diet ($P < 0.05$) (Table 2). The other variables were not affected ($P > 0.10$) by the level or source of lysine. In finishing stage II, the pigs fed 1.03% digestible lysine had the highest ADFI ($P = 0.08$), although the source of lysine did not influence this variable ($P > 0.10$). The final bodyweight was also increased ($P = 0.009$) when feeding 1.03% digestible lysine compared with lower levels, regardless of the source of lysine. The plasma urea nitrogen increased ($P = 0.03$) with 1.03% digestible lysine and was reduced when feeding protected lysine ($P = 0.06$). No effect ($P > 0.10$) of the level or source of lysine was observed on the ADG, FGR, FFLG, backfat thickness and LMA. Thus, the heaviest final bodyweight was observed in all three stage, ADFI was higher in the growing and finishing stage II, and LMA and lean meat percentage were greatest in finishing stage I when the concentration of dietary digestible lysine was greater than the recommendations of the NRC (2012) and Brazilian tables (Rostagno *et al.*, 2017).

Too low a level of dietary lysine compromises the growth of pigs. The lack of lysine, as a constituent of protein, could be the primary reason for its reduced and consequently compromised FGR and ADG (Hasan *et al.*, 2020). There is conflicting information about the performance of pigs when the digestible lysine level is increased in their diets. Increasing lysine levels above NRC (2012) and Brazilian tables (Rostagno *et al.*, 2017) recommendations improved some growth performance variables (backfat thickness, LMA, lean meat percentage, FFLG, ADG and ADFI) (Figueroa *et al.*, 2012; Zhang *et al.*, 2013; Goncalves *et al.*, 2017). Ma *et al.* (2015) also found that higher dietary lysine concentration improved ADG, FGR and FFLG. The results of the present trial agree closely with those of Shelton *et al.* (2011) in growing and finishing I pigs, and with those obtained by Webster *et al.* (2007) in finishing II phase when ractopamine was fed at 5 ppm. In a meta-analysis (Goncalves *et al.*, 2017) concluded that the digestible lysine requirements for barrows, gilts and boars, after adjusting for feed intake, were 106%, 103%, and 132% of the NRC (2012) recommendations, respectively. However, in other studies (Cu *et al.*, 2012; Rivero *et al.*, 2013; Gutiérrez-Hernández *et al.*, 2016), no effect was observed for an increased concentration of dietary lysine on growth and carcass characteristics. This inconsistency in reports about higher levels of lysine in fattening pig diets on growth may be because of different productive conditions, genetic potential of pigs, and level of inclusion (Palma-Granados *et al.*, 2019; Soto *et al.*, 2019; Remus *et al.*, 2020).

Supplementing the diets for finishing pigs in stage III with protected lysine produced no effect in the present trial (Table 3). However, in the growing stage, protected lysine altered ADFI, ADG, final bodyweight, FFLG and LMA. The seemingly conflicting results might be attributed to the lower ability of smaller pigs to digest the protective layer resulting in less availability and absorption of lysine. However, Prandini *et al.* (2013) observed better metabolic efficiency for protected amino acids compared with their conventional counterparts when used in pig feed. In laying hens and broilers, there were indications that the addition of protected lysine and methionine reduced the post-absorptive concentration of these amino acids, improving bio-efficiency and contributing to reduced dietary requirements for these nutrients (Sun *et al.*, 2020a, b).

Table 1 Performance of pigs fed diets that contained three levels lysine in either the protected or conventional form during the growing phase

Lysine level, %	Form	ADFI, kg d ⁻¹	ADG, kg d ⁻¹	FGR	FBW, kg	FFLG, kg d ⁻¹	LMP, %	BT, mm	LMA, cm ²	PUN, mg dL ⁻¹
0.90	Conventional	1.99 ^{ab}	0.68	2.91	54.71 ^{ab}	0.25	29.01	9.29	21.50	21.64
0.90	Protected	1.76 ^a	0.66	2.75	53.18 ^a	0.23	28.71	8.63	19.69	21.98
1.00	Conventional	2.10 ^{ab}	0.74	2.83	57.71 ^b	0.27	28.77	9.14	21.49	23.21
1.00	Protected	1.88 ^{ab}	0.68	2.78	54.92 ^{ab}	0.24	28.96	9.33	21.29	19.15
1.10	Conventional	2.16 ^b	0.73	2.94	56.26 ^{ab}	0.26	29.52	9.13	22.46	22.05
1.10	Protected	2.13 ^{ab}	0.68	3.07	53.79 ^{ab}	0.24	28.88	8.86	20.39	21.41
SE		0.09	0.03	0.13	1.11	0.013	0.35	0.29	0.78	2.63
0.90		1.86 ^a	0.67	2.82	53.90 ^a	0.24	28.85	8.93	20.53	21.83
1.00		2.00 ^{ab}	0.70	2.80	56.42 ^b	0.25	28.86	9.23	21.40	21.18
1.10		2.15 ^b	0.71	3.00	55.10 ^{ab}	0.25	29.22	9.00	21.49	21.77
SE		0.07	0.02	0.09	0.79	0.01	0.24	0.21	55.39	1.86
		Conventional	2.08 ^a	0.71 ^a	2.89	56.23 ^a	0.26 ^a	29.11	9.18	21.84 ^a
		Protected	1.91 ^b	0.67 ^b	2.86	53.88 ^b	0.23 ^b	28.84	8.90	20.38 ^b
SE		0.05	0.03	0.03	0.64	0.03	0.20	0.16	0.44	1.51
<i>P</i> -values										
Level		0.02	0.29	0.30	0.09	0.50	0.49	0.58	0.39	0.96
Form		0.07	0.10	0.82	0.01	0.05	0.36	0.27	0.03	0.53
Interaction		0.05	0.35	0.58	0.06	0.38	0.61	0.50	0.16	0.93

ADFI: average daily feed intake, ADG: average daily gain, FGR: feed to gain ratio, FBW: final bodyweight, FFLG: Fat free lean gain, LMP: lean meat percentage, BT: backfat thickness, LMA: *longissimus* muscle area, PUN: plasma urea nitrogen concentration

^{a,b} Within a column and effect, means with a common superscript were not different with probability *P* ≤ 0.1

Table 2 Performance of pigs fed diets that contained three levels lysine in protected or conventional forms during finishing I phase

Lysine level, %	Form	ADFI, kg d ⁻¹	ADG, kg d ⁻¹	FGR	FBW, kg	FFLG, kg d ⁻¹	LMP, %	BT, mm	LMA, cm ²	PUN, mg dL ⁻¹	
0.75	Conventional	2.88	0.84	3.54	75.83 ^a	0.31	28.39	11.83	26.73 ^a	11.78	
0.75	Protected	3.13	0.95	3.36	80.80 ^{ab}	0.33	27.72	10.80	25.81 ^a	11.92	
0.85	Conventional	3.13	0.91	3.46	82.76 ^b	0.34	27.88	10.60	27.02 ^a	9.61	
0.85	Protected	2.77	0.90	3.11	79.86 ^{ab}	0.35	28.84	10.71	28.86 ^{ab}	13.48	
0.95	Conventional	3.32	0.93	3.58	83.42 ^b	0.35	29.07	11.83	31.24 ^b	11.92	
0.95	Protected	3.02	0.92	3.32	81.71 ^{ab}	0.36	28.96	11.00	29.46 ^{ab}	13.27	
SE		0.17	0.06	0.24	1.62	0.01	0.40	0.59	1.06	2.84	
0.75		2.99	0.89	3.46	78.1 ^a	0.32	28.09 ^a	10.82	26.31 ^a	11.85	
0.85		2.92	0.90	3.25	81.1 ^{ab}	0.34	28.47 ^{ab}	10.67	28.09 ^{ab}	11.87	
0.95		3.16	0.92	3.44	82.5 ^b	0.35	29.01 ^b	11.38	30.28 ^b	13.74	
SE		0.12	0.04	0.18	1.15	0.01	0.28	0.42	75.29	2.01	
		Conventional	3.11	0.89	3.52	80.54	0.33	28.61	11.12	28.41	12.03
		Protected	2.96	0.92	3.25	80.79	0.35	28.48	10.84	28.28	12.99
SE		0.10	0.03	0.14	0.94	0.01	0.23	0.35	0.62	1.65	
P-value											
Level		0.33	0.77	0.65	0.03	0.23	0.07	0.42	0.003	0.73	
Form		0.27	0.62	0.19	0.97	0.32	0.79	0.56	0.70	0.68	
Interaction		0.22	0.80	0.74	0.03	0.53	0.12	0.72	0.10	0.88	

ADFI: average daily feed intake, ADG: average daily gain, FGR: feed to gain ratio, FBW: final bodyweight, FFLG: Fat free lean gain, LMP: lean meat percentage, BT: backfat thickness, LMA: *longissimus* muscle area, PUN: plasma urea nitrogen concentration

^{a,b} Within a column and effect, means with a common superscript were not different with probability $P \leq 0.1$

Table 3 Performance of pigs fed diets that contained three levels lysine in protected or conventional forms during finishing II phase

Lysine level, %	Form	ADFI, kg d ⁻¹	ADG, kg d ⁻¹	FGR	FBW, kg	FFLG, kg d ⁻¹	LMP, %	BT, mm	LMA. cm ²	PUN, mg dL ⁻¹
0.83	Conventional	2.84	0.84	3.48	96.9 ^a	0.31	27.82	14.50	32.90	12.84
0.83	Protected	3.22	0.96	3.42	104.0 ^{ab}	0.32	26.73	15.29	31.11	10.16
0.93	Conventional	3.13	0.88	3.57	104.9 ^{ab}	0.26	26.81	15.20	31.00	12.09
0.93	Protected	2.79	0.84	3.33	100.9 ^{ab}	0.29	27.58	15.33	33.49	10.47
1.03	Conventional	3.48	0.85	4.10	106.5 ^b	0.29	28.05	15.40	36.51	14.22
1.03	Protected	3.31	0.91	3.67	108.2 ^b	0.31	27.06	16.43	31.55	13.66
SE		0.21	0.06	0.24	2.18	0.02	0.36	0.87	3.16	1.08
		3.03 ^{ab}	0.90	3.44	100.7 ^a	0.31	27.23	14.92	31.93	11.40 ^a
		2.94 ^a	0.86	3.44	102.7 ^a	0.27	27.23	15.27	32.36	11.21 ^a
		3.38 ^b	0.89	3.85	107.5 ^b	0.30	27.82	16.00	33.61	13.90 ^b
	SE	0.14	0.04	0.17	1.56	0.02	0.25	0.62	2.26	0.78
SE	Conventional	3.13	0.86	3.70	102.41	0.29	25.57	15.00	33.43	13.04 ^a
	Protected	3.12	0.90	3.48	104.61	0.30	27.31	15.70	31.97	11.48 ^b
	SE	0.01	0.06	0.14	1.26	0.01	0.21	0.50	1.83	0.63
<i>P</i> -value										
Level		0.08	0.81	0.14	0.009	0.40	0.16	0.43	0.85	0.03
Form		0.86	0.34	0.22	0.28	0.40	0.32	0.34	0.55	0.06
Interaction		0.15	0.73	0.31	0.009	0.75	0.11	0.71	0.84	0.11

ADFI: average daily feed intake, ADG: average daily gain, FGR: feed to gain ratio, FBW: final bodyweight, FFLG: fat free lean gain, LMP: lean meat percentage, BT: backfat thickness, LMA: *longissimus* muscle area, PUN: plasma urea nitrogen concentration

^{a,b} Within a column and effect, means with a common superscript were not different with probability *P* ≤ 0.1

In finishing II stage, the plasma urea nitrogen was reduced in pigs fed with protected lysine and increased with the highest concentration of lysine. However, in the other stages, there was no effect of lysine level and source on this variable. The lower values of plasma urea nitrogen with protected lysine confirm its lower bio-efficiency and availability and lower plasma urea nitrogen was not reflected in higher ADG and lean meat percentage. Although other studies reported that the increase of lysine levels minimized plasma urea nitrogen (Zhang *et al.*, 2011; Martínez-Aispuro *et al.*, 2014; Ma *et al.*, 2015), in this study, higher lysine level increased plasma urea nitrogen, possibly because of its relationship with other amino acids, and since this was not constant when increasing lysine, an imbalance between them could be produced (Abreu *et al.*, 2007ab). Hasan *et al.* (2017) observed that a dietary lysine restriction in pigs is responsible for reduced plasma lysine, methionine, leucine, arginine, tyrosine, and total protein, and growth performance. In addition, higher ADFI combined with a high concentration of lysine could cause the plasma urea nitrogen to be elevated. In growing and finishing I stages of the present study there was no effect on plasma urea nitrogen that would indicate an adequate balance of amino acids (Qin *et al.*, 2015; Figueroa-Velasco *et al.*, 2020).

Conclusion

The lysine level that is needed in the diets of pigs may be 0.10% higher than recommended levels. The use of protected lysine did not improve the growth performance and carcass characteristics of finishing pigs. Further, the use of protected lysine affected some growth performance variables in growing pigs in a negative way. In addition, the lower values of plasma urea nitrogen observed with protected lysine could indicate that this type of amino acid has lower bioefficiency and availability.

Authors' Contributions

JLFV, DTSL, JAMA, TSTE, JLKM, ARF, MMCG were responsible for the design and execution of the proposal research. All co-authors participated in the management and discussion of the results, statistical analysis and writing of the manuscript.

Conflict of Interest Declaration

Authors declare that there is no conflict of interest regarding the publication of this article.

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Supplemental Table S1 Composition of experimental diets for growing pigs fed with three levels and two types of lysine

Ingredient, %	Dietary treatment					
	1	2	3	4	5	6
Sorghum grain	77.88	77.69	77.62	77.35	77.35	77.35
Soybean meal	18.57	18.59	18.61	18.61	18.64	18.64
Soybean oil	0.93	0.96	0.97	1.00	1.00	1.04
Bio lysine ¹	0.54	0.00	0.74	0.00	0.93	0.00
DL-Methionine	0.16	0.16	0.16	0.16	0.16	0.16
L-Threonine	0.10	0.10	0.10	0.10	0.10	0.10
Vitamins ²	0.18	0.18	0.18	0.18	0.18	0.18
Minerals ³	0.18	0.18	0.18	0.18	0.18	0.18
Protected lysine	0.00	0.68	0.00	0.93	0.00	1.18
Common salt	0.30	0.30	0.30	0.30	0.30	0.30
Calcium carbonate	0.91	0.91	0.91	0.91	0.911	0.91
Orthophosphate	0.26	0.26	0.26	0.26	0.26	0.26
Calculated nutrient composition, %						
ME, Mcal Kg ⁻¹	3.30	3.30	3.30	3.30	3.30	3.30
Crude protein	16.47	16.56	16.59	16.70	16.70	16.84
Calcium	0.67	0.67	0.67	0.67	0.67	0.67
Phosphorus	0.31	0.31	0.31	0.31	0.31	0.31
Digestible lysine	0.90	0.90	1.00	1.00	1.10	1.10
Digestible threonine	0.60	0.60	0.60	0.60	0.60	0.60
Digestible tryptophan	0.17	0.17	0.17	0.17	0.17	0.17
Digestible methionine	0.38	0.38	0.38	0.38	0.38	0.38
Digestible methionine+cysteine	0.57	0.57	0.57	0.57	0.57	0.57
Measured nutrient composition, %						
Crude protein	16.75	15.86	15.88	16.71	15.90	15.95
Calcium	0.62	0.65	0.63	0.64	0.63	0.66
Phosphorus	0.30	0.28	0.28	0.27	0.29	0.30

¹Bio lysine: 50.7% lysine²(per kg feed) vitamin A: 15000 IU, vitamin D3: 2500 IU, vitamin E: 37.5 IU, vitamin K: 2.5 mg, thiamine: 2.25 mg, riboflavin: 6.25 mg, pyridoxine: 2 mg, niacin: 50 mg, D-calcium pantothenate: 20 mg, folic acid: 1.25 mg, choline chloride: 563 mg, cyanocobalamin: 0.0375 mg, biotin: 0.13 mg³(per kg of feed): Selenium: 0.15 mg, Chromium: 0.2 mg, Iodine: 0.9 mg, Copper: 10 mg, Zinc: 150 mg, Iron: 150 mg, Manganese: 150 mg

Supplemental Table S2 Composition of experimental diets for finishing I pigs fed with three levels and two types of lysine

Ingredient, %	Dietary treatment					
	1	2	3	4	5	6
Sorghum grain	82.13	81.97	81.86	81.63	81.96	81.29
Soybean meal	14.53	14.57	14.57	14.57	14.60	14.65
Soybean oil	0.85	0.87	0.89	0.92	0.92	0.96
Bio lysine*	0.45	0.00	0.64	0.00	0.84	0.00
DL-Methionine	0.10	0.10	0.10	0.10	0.10	0.10
L-Threonine	0.07	0.07	0.07	0.07	0.07	0.07
Vitamins**	0.18	0.18	0.18	0.18	0.18	0.18
Minerals***	0.18	0.18	0.18	0.18	0.18	0.18
Protected lysine	0.00	0.57	0.00	0.81	0.00	1.06
Common salt	0.30	0.30	0.30	0.30	0.30	0.30
Calcium carbonate	0.87	0.87	0.87	0.87	0.87	0.87
Orthophosphate	0.34	0.34	0.34	0.35	0.35	0.35
Nutrient composition calculated, %						
ME, Mcal Kg ⁻¹	3.30	3.30	3.30	3.30	3.30	3.30
Crude protein	14.84	14.91	14.95	15.05	15.06	15.19
Calcium	0.67	0.67	0.67	0.67	0.67	0.67
Phosphorus	0.31	0.31	0.31	0.31	0.31	0.31
Digestible lysine	0.75	0.75	0.85	0.85	0.95	0.95
Digestible threonine	0.52	0.52	0.52	0.52	0.52	0.52
Digestible tryptophan	0.15	0.15	0.15	0.15	0.15	0.15
Digestible methionine	0.31	0.31	0.31	0.31	0.31	0.31
Digestible methionine+cCysteine	0.48	0.48	0.48	0.48	0.48	0.48
Nutrient composition determined, %						
Crude protein	14.78	14.86	14.88	14.92	14.89	14.99
Calcium	0.63	0.61	0.64	0.65	0.66	0.64
Phosphorus	0.30	0.28	0.28	0.27	0.29	0.30

*Bio lysine: 50.7% lysine

²(per kg feed) vitamin A: 15000 IU, vitamin D3: 2500 IU, vitamin E: 37.5 IU, vitamin K: 2.5 mg, thiamine: 2.25 mg, riboflavin: 6.25 mg, pyridoxine: 2 mg, niacin: 50 mg, D-calcium pantothenate: 20 mg, folic acid: 1.25 mg, choline chloride: 563 mg, cyanocobalamin: 0.0375 mg, biotin: 0.13 mg

³Per kg of feed: selenium: 0.15 mg, chromium: 0.2 mg, iodine: 0.9 mg, copper: 10 mg, zinc: 150 mg, iron: 150 mg, manganese: 150 mg

Supplemental Table S3 Composition of experimental diets for finishing II pigs fed with three levels and two types of lysine

Ingredient, %	Dietary treatment					
	1	2	3	4	5	6
Sorghum grain	80.13	79.95	79.86	79.61	79.59	79.27
Soybean meal	16.53	16.56	16.57	16.61	16.61	16.65
Soybean oil	0.84	0.860	0.87	0.91	0.91	0.95
Bio lysine ¹	0.50	0.00	0.70	0.00	0.90	0.0
DL-Methionine	0.13	0.13	0.13	0.13	0.13	0.13
L-Threonine	0.10	0.10	0.10	0.10	0.10	0.10
Vitamins ²	0.18	0.18	0.18	0.18	0.18	0.18
Minerals ³	0.18	0.18	0.18	0.18	0.18	0.18
Protected lysine	0.00	0.64	0.00	0.89	0.00	1.13
Common salt	0.30	0.30	0.30	0.30	0.30	0.30
Calcium carbonate	0.81	0.81	0.81	0.81	0.81	0.81
Orthophosphate	0.30	0.30	0.30	0.30	0.30	0.30
Nutrient composition calculated, %						
ME, Mcal Kg ⁻¹	3.30	3.30	3.30	3.30	3.30	3.30
Crude protein	15.68	15.75	15.79	15.89	15.90	16.03
Calcium	0.64	0.64	0.64	0.64	0.64	0.64
Phosphorus	0.31	0.31	0.31	0.31	0.31	0.31
Digestible lysine	0.83	0.83	0.93	0.93	1.03	1.03
Digestible threonine	0.57	0.57	0.57	0.57	0.57	0.57
Digestible tryptophan	0.16	0.16	0.16	0.16	0.16	0.16
Digestible methionine	0.35	0.35	0.35	0.35	0.35	0.35
Digestible methionine+cysteine	0.53	0.53	0.53	0.53	0.53	0.53
Nutrient composition determined, %						
Crude protein	14.95	15.60	15.40	15.81	15.70	15.90
Calcium	0.64	0.63	0.62	0.61	0.64	0.60
Phosphorus	0.32	0.30	0.31	0.29	0.31	0.30

¹Bio lysine: 50.7% lysine²Per kg feed: vitamin A: 15000 IU, vitamin D3: 2500 IU, vitamin E: 37.5 IU, vitamin K: 2.5 mg, thiamine: 2.25 mg, riboflavin: 6.25 mg, pyridoxine: 2 mg, niacin: 50 mg, D-calcium pantothenate: 20 mg, folic acid: 1.25 mg, choline chloride: 563 mg, cyanocobalamin: 0.0375 mg, biotin: 0.13 mg³Per kg of feed: selenium: 0.15 mg, chromium: 0.2 mg, iodine: 0.9 mg, copper: 10 mg, zinc: 150 mg, iron: 150 mg, manganese: 150 mg