Modeling the effects of standardized ileal digestible isoleucine to lysine ratio on growth performance of nursery pigs[1,2]

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ABSTRACT: Two experiments evaluated the effects of increasing standardized ileal digestible (SID) Ile:Lys ratio on growth performance of nursery pigs. In both experiments, dietary treatments consisted of 40, 44, 48, 52, 54, 58, or 63% SID Ile:Lys ratio. Diets were formulated using analyzed ingredient AA values and NRC (2012) SID coefficients. A combination of field peas and spray dried blood cells were used to ensure a low enough Ile diet concentration while minimizing the excess of Leu. The experiments consisted of 8 pens per dietary treatment with 5 pigs per pen for a total of 280 nursery pigs per experiment (Exp. 1: PIC 327 × 1,050, initially 6.7 ± 1.0 kg BW; Exp. 2: DNA 600 × 241, initially 6.0 ± 0.97 kg BW). Data were analyzed using mixed models with heterogeneous variance, where appropriate. The dose response was further characterized using quadratic polynomial (QP), broken-line linear (BLL), or broken-line quadratic (BLQ) functional forms. For Exp. 1, diets were initiated 6-d post-weaning and fed for 12-d followed by a common diet from d 12 to 28. From d 0 to 12, increasing dietary SID Ile:Lys ratio increased ADG (linear, $P < 0.005$) and ADFI (quadratic, $P < 0.017$) but G:F decreased (quadratic, $P < 0.043$). For ADG, the QP, BLL, and BLQ models resulted in maximum ADG at 64.7, 52.0, and 52.0 SID Ile:Lys ratios, respectively. For ADFI, the BLL breakpoint occurred at 50.6 and the QP predicted maximum ADFI at 56.2 SID Ile:Lys ratio. In Exp. 2, diets were initiated 6-d post-weaning for 7 pens and 3-d post-weaning for one heavier block and fed for 18-d followed by a common diet from d 18 to 32. From d 0 to 18, ADG and ADFI increased (quadratic, $P < 0.016$) with no evidence for difference in G:F as SID Ile:Lys ratio increased. For ADG, the QP and BLL had similar fit with breakpoints or maximums occurring at 58.3 and 51.8% SID Ile:Lys ratio, respectively. For ADFI, the BLQ breakpoint occurred at 52.0 SID Ile:Lys and the QP maximum ADFI at 57.2% SID Ile:Lys ratio. In conclusion, broken-line models reported maxima of 52.0% Ile:Lys ratio while quadratic models were as high as 64% of Lys to maximize ADG and ADFI of 6- to 11-kg nursery pigs. However, for the QP models 99% of the maximum response was achieved with a dose comparable to that from the broken line models. Therefore, these results are similar to the NRC (2012) requirement estimate of 51.1 Ile:Lys ratio.

Key words: amino acids, growth, isoleucine, nursery, pig

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INTRODUCTION

Diets for nursery pigs can be supplemented with crystalline Lys, Met, Thr, Trp, Val without loss of performance (Nørgaard and Fernández, 2009; Gloaguen et al., 2014). After these 5 amino acids, Ile is the next limiting amino acid in many practical formulations.

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Thus, more trials are needed to further characterize the Ile requirement under practical conditions.

Research evaluating Ile requirements in nursery pigs usually takes one of two approaches, using either high dietary levels of spray dried blood cells (SDBC), which contain low Ile and high Leu concentrations, or diets without SDBC. In diets containing SDBC, excess Leu can be problematic as it increases production of branched-chain keto-acid dehydrogenase, which in turn metabolizes all branched chain amino acids (BCAA), causing an increased catabolism of Ile and raising the actual requirement (Langer et al., 2000; Wiltafsky et al., 2010; Morales et al., 2016).

Using the second approach of studies using diets without added blood cells, Ile may not have been low enough (NRC, 2012) to elicit a response or other AA could have been limiting (van Milgen et al., 2012). One approach to ensure a low enough dose to elicit a dose response is to use an ingredient that is both low in Ile and Leu relative to the other supplemented amino acids. One ingredient that fits this profile and is commonly used in practical diet formulation around the world is field peas. Therefore, in this experiment, field peas and low levels of SDBC were used to achieve low dietary Ile without excess Leu. Thus, the objective was to assess the Ile requirement for growth performance of nursery pigs using diets without excessive Leu.

MATERIALS AND METHODS

The Kansas State University Institutional Animal Care and Use Committee approved the protocol used in the 2 experiments reported herein.

General

Both experiments were conducted at the Kansas State University Swine Teaching and Research Center using similar protocols, described as follows. Each pen (1.52 × 1.52 m, Exp. 1; 1.52 × 1.22 m, Exp. 2) contained a 4-hole dry self-feeder and a nipple waterer for ad libitum access to feed and water.

Dietary treatments were corn- and soybean meal-based containing 10% dried whey, 10% field peas, and 1.5% SDBC. Corn, soybean meal, field peas, and dried whey were analyzed for total AA content (excluding Trp; method 994.12; AOAC International, 2012), Trp (method 13904:2005; ISO, 2005), and CP (method 990.03; AOAC International, 2012) by Ajinomoto Heartland, Inc. (Chicago, IL) prior to formulation (Table 1). A new

Table 1. Chemical analysis of ingredients

Item, %	Corn[1,2]	Soybean meal[1,2]	Dried whey[1,2]	Field peas, Exp. 1[3]	Field peas, Exp. 2[1,3]	Spray dried blood cells[4]
DM	88.81	89.09	90.04	92.19	86.60	93.30
CP	9.013	47.00	11.84	22.11	22.50	92.60
Total AA						
Lys	0.29	2.88	0.79	1.54	1.59	8.88
Ile	0.31	2.09	0.65	0.89	0.91	0.28
Leu	1.10	3.51	1.07	1.52	1.59	12.62
Met	0.18	0.66	0.16	0.19	0.21	1.23
Thr	0.30	1.80	0.68	0.79	0.83	4.29
Trp	0.07	0.65	0.22	0.17	0.21	1.58
Val	0.40	2.12	0.59	0.99	0.99	8.35
His	0.24	1.17	0.18	0.50	0.54	6.17
Phe	0.43	2.35	0.37	1.03	1.07	7.25
Standardized ileal digestible AA, % (Calculated)						
Lys	0.21	2.56	0.77	1.40	1.44	8.67
Ile	0.25	1.86	0.62	0.78	0.79	0.25
Leu	0.95	3.09	1.05	1.35	1.41	12.3
Met	0.15	0.59	0.16	0.17	0.19	1.26
Thr	0.23	1.53	0.61	0.69	0.72	4.13
Trp	0.06	0.59	0.21	0.15	0.18	1.48
Val	0.32	1.84	0.56	0.86	0.86	8.14
His	0.20	1.05	0.17	0.46	0.50	6.07
Phe	0.36	2.07	0.34	0.92	0.96	7.08

[1]Analyzed at Ajinomoto Heartland, Inc. (Chicago, IL) for amino acid content.

[2]SID content calculated using SID coefficients from the NRC (NRC. 2012).

[3]Exp. 1 peas used total AA content and SID coefficients from Mathai (2015). Exp. 2 peas use SID coefficients from Mathai (2015).

[4]Spray dried blood cells use total values and coefficients from Almeida et al. (2013).

field pea batch and analysis prior to Exp. 2 called for a minor adjustment to Exp. 2 diets by decreasing crystalline amino acids slightly. The standardized ileal digestibility (SID) coefficients were obtained from NRC for all ingredients except field peas which were from Mathai (2015). Crystalline amino acids replaced corn in diets as treatment levels of Ile increased.

Except for Lys and Ile, all other AA were formulated above their requirement estimates (NRC, 2012). Based on a previous study by Clark et al. (2016), the Lys requirement for nursery pigs of this weight range in these facilities was at least 1.45% SID Lys. Thus, experimental diets were formulated to contain 1.28 and 1.24% SID Lys for Exp. 1 and 2, respectively, thus ensuring that pigs were below their Lys requirement and guaranteeing that Lys was second limiting. Dietary treatments consisted of 40, 44, 48, 52, 54, 58, and 63% SID Ile:Lys ratio. Basal diets were manufactured for the lowest and highest Ile:Lys ratio treatments and then blended at the feed mill to create the intermediate levels. After the experimental period, a common-phase diet was fed, consisting of a corn- and soybean meal- base, containing no animal protein sources and formulated to 1.22% SID Lys. All diets were fed in meal form and prepared at the O.H. Kruse Feed Technology Innovation Center located in Manhattan, KS. Samples of experimental diets were submitted (Ward Laboratories, Kearney, NE) for analysis of DM (method 935.29; AOAC International, 2012), CP (method 990.03; AOAC International, 2012), crude fiber [method 978.10; AOAC International, 2012 for preparation and Ankom 2000 Fiber Analyzer (Ankom Technology, Fairport, NY)], ash (method 942.05; AOAC International, 2012), and ether extract [method 920.39 a; AOAC International, 2012 for preparation and ANKOM XT20 Fat Analyzer (Ankom Technology). Samples were also analyzed for AA analysis using methods described above (Ajinomoto Heartland, Inc., Chicago, IL).

Experiment 1

A total of 280 nursery pigs (PIC 327 × 1,050; 6.7 ± 1.0 kg BW) were used in a 26-d experiment with 8 pens per dietary treatment and 5 pigs per pen. Pigs were weaned at approximately 21 d of age and allotted to the nursery according to BW. After 6 d in the nursery, pens were allotted to dietary treatments in a randomized complete block design using BW blocks. Dietary treatments were fed for 12 d followed by a common-phase diet for additional 14 d (Table 2). Pens of pigs were weighed and feed disappearance was measured on d 0, 12, and 26 of the study.

Table 2. Diet composition (Exp. 1, as-fed basis)[1]

Item	Formulated SID[2] Ile:Lys ratio, % 40	63	Common phase
Ingredient, %			
Corn	57.68	57.59	63.77
Soybean meal, 48% CP	13.25	13.26	32.86
Dried whey	10.00	10.00	–
Field peas	10.00	10.00	–
Spray dried blood cells	1.50	1.50	–
Limestone	1.00	1.00	0.98
Monocalcium phosphate, 22% P	1.80	1.80	1.10
Salt	0.30	0.30	0.35
L-Lys-HCl	0.63	0.63	0.30
DL-Met	0.33	0.33	0.12
L-Thr	0.32	0.32	0.12
L-Trp	0.10	0.10	–
L-Val	0.24	0.24	–
L-Ile	–	0.29	–
Glutamic acid	1.10	1.00	–
Glycine	1.10	1.00	–
Trace mineral premix[3]	0.15	0.15	0.15
Vitamin premix[4]	0.25	0.25	0.25
Zinc oxide	0.25	0.25	–
Total	100.00	100.00	100.00
Calculated analysis			
SID AA, %			
Lys	1.28	1.28	1.22
Ile:Lys	40	63	63
Leu:Lys	107	107	129
Met:Lys	42	42	33
Met and Cys:Lys	59	59	57
Thr:Lys	65	65	63
Trp:Lys	20.3	20.3	18.7
Val:Lys	71	71	69
Total Lys, %	1.38	1.38	1.37
ME, kcal/kg	3,228	3,236	3,272
NE, kcal/kg	2,427	2,436	2,407
SID Lys:ME, g/Mcal	3.96	3.95	3.73
SID Lys:NE, g/Mcal	5.36	5.34	5.16
CP, %	18.2	18.2	21.4
Ca, %	0.82	0.82	0.70
P, %	0.73	0.73	0.64
Available P, %	0.51	0.51	0.41

[1]Dietary treatments 40% and 63% SID Ile:Lys were manufactured and blended at the feed mill to create the intermediate levels of 44, 48, 52, 54, and 58% SID Ile:Lys.

[2]Standardized ileal digestible.

[3]Provided per kilogram of premix: 22 g Mn from manganese oxide; 73 g Fe from iron sulfate; 73 g Zn from zinc sulphate; 11 g Cu from copper sulfate; 198 mg I from calcium iodate; and 198 mg Se from sodium selenite.

[4]Provided per kilogram of premix: 3,527,360 IU vitamin A; 881,840 IU vitamin D3; 17,637 IU vitamin E; 3,307 mg riboflavin; 1,764 mg menadione; 11,023 mg pantothenic acid; 33,069 mg niacin; and 15.4 mg vitamin B12.

Experiment 2

A total of 280 nursery pigs (DNA 600 × 241, initially 6.0 ± 0.97 kg BW) were used in a 32-d experiment with 8 pens per dietary treatment and 5 pigs per pen. Pigs were weaned at approximately 20 d of age and randomly allotted to pens in blocks according to BW in a randomized complete block design. Dietary treatments were fed for 18 d followed by a common diet for 14 d (Table 3). Pens of pigs were weighed and feed disappearance was measured on d 0, 18, and 32 of the study.

Statistical Analysis

Statistical analyses for these experiments were performed using methods described by Gonçalves et al. (2016). Models were evaluated separately for each experiment. Briefly, for each experiment, we first fitted a base mixed model including the fixed effect of dietary treatment and the random effect block with pen as the experimental unit. The model was also used to explore heterogeneity of residual variances across dietary treatments and Bayesian Information Criteria (BIC) was used to decide on the best fitting model to account for heteroskedasticity. For Exp. 1, heterogeneous variance was applied for both ADG (40, 48, 52, and 54% Ile:Lys dietary treatments vs. 44, 58, and 63% SID Ile:Lys ratio dietary treatments) and G:F (54% SID Ile:Lys vs. all other dietary treatments) during the experimental period. For Exp. 2, heterogeneous variance was applied for ADG (48% Ile:Lys ratio dietary treatment vs. all other dietary treatments) and ADFI (48% Ile:Lys ratio dietary treatment vs. all other dietary treatments). After accounting for heterogeneous variance, orthogonal linear and quadratic polynomial contrasts accounting for unequal spacing between dietary treatments were evaluated.

The dose response was further assessed in both experiments using continuous response models fit to ADG and ADFI during the experimental periods (Exp. 1: d 0 to 12; Exp. 2: d 0 to 18). These procedures evaluated the functional forms of the relationship between ADG or ADFI and dietary treatments. The competing models evaluated were the quadratic polynomial (QP), broken-line linear (BLL), or broken-line quadratic (BLQ), following Gonçalves et al. (2016). The best-fitting dose–response model was decided using BIC, whereby decreases of 2 points or more to indicate evidence for enhanced fit of the model with lower BIC. These guidelines were based on the suggestions of Raftery (1996) and Gonçalves et al. (2016).

For each best fitting dose–response model, the individual pen means and fitted values were plotted. For the BLL and BLQ models, the estimated breakpoints with the respective 95% CI were reported. For the QP model the estimated SID % at maximum response and

Table 3. Diet composition (Exp. 2, as-fed basis)[1]

	Formulated SID[2] Ile:Lys ratio, %		Common
Item	50	85	phase
Ingredient, %			
Corn	59.04	58.95	63.77
Soybean meal, 48% CP	11.95	11.96	32.86
Dried whey	10.00	10.00	–
Field peas	10.00	10.00	–
Spray dried blood cells	1.50	1.50	–
Limestone	1.00	1.00	0.98
Monocalcium phosphate, 22% P	1.80	1.80	1.10
Salt	0.30	0.30	0.35
L-Lys-HCl	0.60	0.60	0.30
DL-Met	0.32	0.32	0.12
L-Thr	0.31	0.31	0.12
L-Trp	0.10	0.10	–
L-Val	0.23	0.23	–
L-Ile	–	0.28	–
Glutamic acid	1.10	1.00	–
Glycine	1.10	1.00	–
Trace mineral premix[3]	0.15	0.15	0.15
Vitamin premix[4]	0.25	0.25	0.25
Zinc oxide	0.25	0.25	–
Total	100.00	100.00	100.00
Calculated analysis			
SID AA, %			
Lys	1.24	1.24	1.22
Ile:Lys	40	63	63
Leu:Lys	109	109	129
Met:Lys	42	42	33
Met and Cys:Lys	60	60	57
Thr:Lys	66	66	63
Trp:Lys	21	21	18.7
Val:Lys	71	71	69
Total Lys, %	1.34	1.34	1.37
ME, kcal/kg	3,228	3,236	3,272
NE, kcal/kg	2,434	2,443	2,407
SID Lys:ME, g/Mcal	3.83	3.82	3.73
SID Lys:NE, g/Mcal	5.15	5.14	5.16
CP, %	18.6	18.6	21.4
Ca, %	0.81	0.81	0.70
P, %	0.73	0.73	0.64
Available P, %	0.51	0.51	0.41

[1]Dietary treatments 40% and 63% SID Ile:Lys were manufactured and blended at the feed mill to create the intermediate levels of 44, 48, 52, 54, and 58% SID Ile:Lys.

[2]Standardized ileal digestible

[3]Provided per kilogram of premix: 22 g Mn from manganese oxide; 73 g Fe from iron sulfate; 73 g Zn from zinc sulphate; 11 g Cu from copper sulfate; 198 mg I from calcium iodate; and 198 mg Se from sodium selenite.

[4]Provided per kilogram of premix: 3,527,360 IU vitamin A; 881,840 IU vitamin D3; 17,637 IU vitamin E; 3,307 mg riboflavin; 1,764 mg menadione; 11,023 mg pantothenic acid; 33,069 mg niacin; and 15.4 mg vitamin B12.

its corresponding CI was calculated as explained by Gonçalves et al. (2016). Subsequently, evaluation of the base model linear and quadratic contrasts for feed efficiency resulted in quadratic effect in Exp. 1, with the lowest and the highest SID Ile:Lys ratios having the best G:F. Due to the lack of biological explanation of this response, the dose response for G:F was not modeled. Due to a lack of dose response for feed efficiency the dose response curve was not modeled in Exp 2.

Base models were fitted using the GLIMMIX procedure of SAS (version 9.4; SAS Inst. Inc., Cary, NC). Dose–response models: using PROC GLIMMIX and PROC NLMIXED according to procedures of Gonçalves et al. (2016). Results were considered significant at $P \leq 0.05$ and marginally significant at $P < 0.10$.

RESULTS

Chemical Analysis

Amino acid analysis of ingredients resulted in corn generally being slightly higher in AA concentrations as compared to published values (NRC, 2012) whereas soybean meal showed slightly lower AA concentrations (Table 1). Analysis of AA for field peas in Exp. 1 and for Exp. 2 were like expected values.

Proximate analysis of dietary treatments (Tables 4 and 5) generally matched formulated values. The AA analysis of the high and low Ile:Lys ratio diets were consistent with formulated values. Despite blending of the high and low diets, for a few AA analyses, the increase in Ile across dietary treatments was less than expected but within analytical variation. Amino acid analyses of diets were reasonably consistent with diet formulation with Ile increasing across the dietary treatments and other AA remaining relatively constant.

Experiment 1

From d 0 to 12, when dietary treatments were fed, increasing SID Ile:Lys ratio increased ADG (linear, $P < 0.005$) and ADFI (quadratic, $P < 0.017$; Table 6). However, as SID Ile:Lys ratio increased, G:F decreased then increased (quadratic, $P < 0.043$) with the lowest and highest concentrations of 40 and 63% SID Ile:Lys ratio having the best G:F. During the common-diet phase (d 12 to 28), there was no evidence for carryover dietary treatment differences in ADG, ADFI, or G:F. When the overall period was considered, ADG marginally increased (linear, $P < 0.082$) and ADFI increased (linear, $P < 0.011$) due to increasing SID Ile:Lys ratio in diets from d 0 to 12. Similarly, BW was increased (linear, $P < 0.006$) at the end of phase 1, but there was no evidence of dietary treatment differences for final BW at the end of the common diet period.

For ADG (Fig. 1) from d 0 to 12, the QP, BLL, and BLQ had similar competing fits (BIC = 558.3, 556.6, and 557.9, respectively). The BLL and BLQ reported

Table 4. Chemical analysis of diets (Exp. 1, as-fed basis)[1]

	Formulated standardized ileal digestible (SID) Ile:Lys ratio, %[2]						
Item	40	44	48	52	54	58	63
Proximate analysis, %[3]							
DM	88.11	88.82	89.21	88.94	87.85	88.86	89.23
CP	18.0	18.7	18.6	18.7	18.8	18.8	19.0
Crude fiber	2.2	2.0	2.1	2.2	2.3	2.0	2.1
Ether extract	2.6	2.4	2.4	2.6	2.3	2.3	2.1
Ash	4.64	5.52	4.79	5.07	5.3	5.06	5.29
AA analysis, %[4]							
Lys	1.28	1.34	1.40	1.42	1.39	1.45	1.41
Ile	0.59	0.67	0.74	0.74	0.79	0.86	0.93
Leu	1.46	1.50	1.52	1.52	1.53	1.51	1.56
Met	0.46	0.50	0.50	0.53	0.49	0.56	0.55
Met + Cys	0.75	0.79	0.79	0.74	0.76	0.84	0.83
Thr	0.93	0.92	0.89	0.97	0.87	0.91	0.95
Trp	0.24	0.26	0.27	0.28	0.26	0.28	0.29
Val	0.92	0.98	0.97	1.04	1.01	1.04	1.07
His	0.43	0.45	0.45	0.46	0.45	0.45	0.46
Phe	0.79	0.80	0.81	0.81	0.79	0.79	0.81

[1]Dietary treatment samples were collected at the feed mill after manufacturing.

[2]Low (40% SID Ile:Lys) and high (63% SID Ile:Lys) diets were blended at the feed mill to create the intermediate dietary treatments.

[3]Composite samples were submitted to Ward Laboratories (Kearney, NE) for proximate analysis.

[4]Composite samples were submitted to Ajinomoto Heartland Inc. (Chicago, IL) for AA analysis.

Table 5. Chemical analysis of diets (Exp. 2, as-fed basis)[1]

	Formulated standardized ileal digestible (SID) Ile:Lys ratio, %[2]						
Item	40	44	48	52	54	58	63
Item, %[3]							
DM	90.29	90.41	90.07	90.37	90.36	90.30	89.97
CP	18.1	18.4	18.6	18.2	18.3	18.7	18.7
Crude fiber	1.9	1.8	2.4	1.7	1.9	1.9	2.3
Ether extract	2.6	2.7	2.4	2.5	2.6	2.7	2.6
Ash	5.25	5.27	5.12	5.24	5.12	5.40	5.18
AA analysis, %[4]							
Lys	1.33	1.34	1.34	1.36	1.36	1.35	1.33
Ile	0.60	0.60	0.65	0.69	0.75	0.75	0.82
Leu	1.47	1.43	1.45	1.47	1.48	1.47	1.48
Met	0.50	0.54	0.51	0.50	0.52	0.50	0.54
Met + Cys	0.75	0.77	0.73	0.75	0.76	0.77	0.82
Thr	0.86	0.84	0.92	0.87	0.90	0.91	0.98
Trp	0.27	0.27	0.27	0.25	0.27	0.26	0.27
Val	0.97	0.96	0.96	0.95	0.99	1.02	0.99
His	0.46	0.43	0.45	0.43	0.45	0.44	0.45
Phe	0.79	0.77	0.78	0.78	0.78	0.78	0.78

[1]Dietary treatment samples were collected at the feed mill after manufacturing.

[2]Low (40% SID Ile:Lys) and high (63% SID Ile:Lys) diets were blended at the feed mill to create the intermediate dietary treatments.

[3]Composite samples were submitted to Ward Laboratories (Kearney, NE) for analysis.

[4]Composite samples were submitted to Ajinomoto Heartland Inc. (Chicago, IL) for AA analysis.

Table 6. Effects of increasing standardized ileal digestible (SID) Ile:Lys ratio on nursery pig growth performance, Exp. 1[1]

	Formulated SID Ile:Lys ratio, %[2]								Probability, $P <$	
Item	40	44	48	52	54	58	63	SEM	Linear	Quadratic
Treatment period (d 0 to 12)										
ADG, g	330	344	342	388	344	358	375	–[3]	0.005	0.418
ADFI, g	495	524	546	601	522	574	555	16.6	0.002	0.017
G:F	0.669	0.657	0.628	0.648	0.658	0.625	0.676	–[4]	0.904	0.043
Post-treatment period (d 12 to 26)										
ADG, g	554	555	557	553	573	576	545	14.5	0.779	0.325
ADFI, g	851	835	851	859	866	891	854	19.4	0.190	0.588
G:F	0.652	0.665	0.655	0.643	0.662	0.647	0.640	0.0102	0.166	0.429
Overall (d 0 to 26)										
ADG, g	450	458	458	477	467	475	467	11.0	0.082	0.270
ADFI, g	687	692	710	740	707	744	716	15.2	0.011	0.106
G:F	0.657	0.662	0.645	0.645	0.660	0.639	0.652	0.0092	0.337	0.504
BW, kg										
d 0	6.7	6.7	6.7	6.7	6.7	6.7	6.7	0.08	0.995	0.993
d 12	10.7	10.9	10.8	11.4	10.9	11.0	11.2	0.19	0.006	0.536
d 26	18.5	18.6	18.7	19.1	18.9	19.1	18.9	0.33	0.105	0.304

[1]A total of 280 nursery pigs (PIC 327 × 1,050, initially 6.7 ± 0.08 kg BW) were used in a 26-d growth trial with 8 pens per dietary treatment and 5 pigs per pen. Pigs were weaned at approximately 21 d, fed a common starter diet for 6 d post-weaning, then placed on experimental diets. Experimental diets were fed from d 0 to 12 and a common diet was fed from d 12 to 26.

[2]Low (40% SID Ile:Lys) and high (63% SID Ile:Lys) complete dietary treatments were blended upon manufacturing at the feed mill to create the 44, 48, 52, 54, and 58% SID Ile:Lys intermediate dietary treatments.

[3]Heteroskedastic modeling resulted in SEM = 8.8 for 40, 48, 52, and 54% dietary treatments and 14.0 for 44, 58, and 63% SID Ile:Lys ratio dietary treatments.

[4]Heteroskedastic modeling resulted in SEM = 0.0160 for 40, 44, 48, 52, 58, and 63% dietary treatments and 0.0096 for 54% SID Ile:Lys ratio dietary treatment.

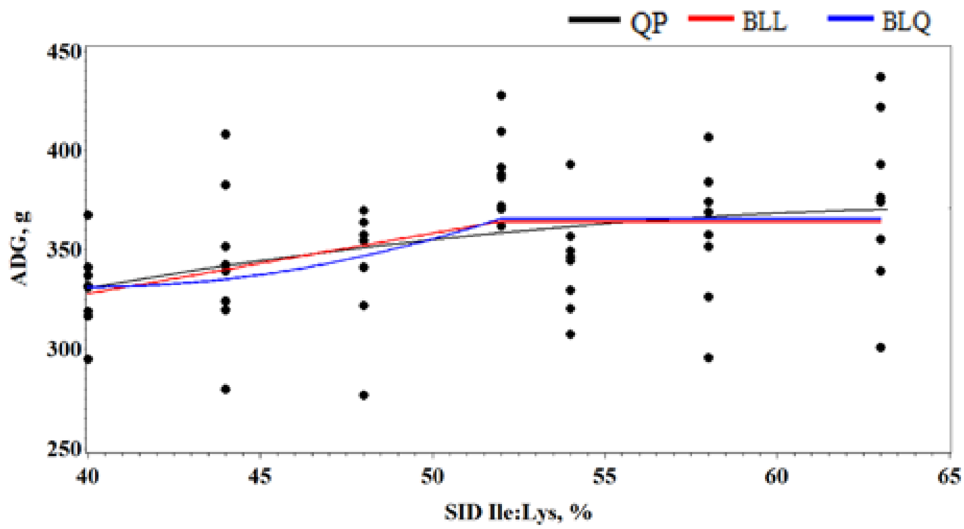

Figure 1. Estimated standardized ileal digestible (SID) Ile:Lys ratio requirement for ADG of nursery pigs, Exp. 1. QP: BIC = 558.3; Estimated 64.7% SID Ile:Lys that maximized ADG; [95% CI: (51, > 65%)]; 99% of Max ADG at 57.0% SID Ile:Lys. BLL: BIC = 556.6; Estimated breakpoint: 52.0% SID Ile:Lys; [95% CI:(51.96, 52.04)]. BLQ: BIC = 557.9; Estimated breakpoint: 52.0% SID Ile:Lys; [95% CI:(51.97, 52.03)]. A total of 280 nursery pigs (PIC 327 × 1,050, initially 6.7 ± 0.08 kg BW) were used in a 26-d growth trial with 5 pigs per pen and 8 pens per treatment. Pigs were weaned at approximately 21 d, fed a common starter diet for 6 d post-weaning, then placed on experimental diets. Experimental diets were fed from d 0 to 12 and a common diet was fed from d 12 to 26. Quadratic polynomial (QP), broken-line linear (BLL), and broken-line quadratic (BLQ) models were fit for the experimental period to estimate SID Ile:Lys. Bayesian Information Criterion (BIC) was used to determine the best fitting models; a lower value indicates a better fit to the data.

similar breakpoints of 52.0% SID Ile:Lys ratio [95% CI: (51.96, 52.04%) and (51.97, 52.03%), respectively]. The QP reported maximum ADG at 64.7% SID Ile:Lys ratio [95% CI: (51, > 65%)] with 99% of maximum ADG captured at a 57.0% SID Ile:Lys ratio.

The estimated regression equation for the BLL model was:

$$\text{ADG, g} = 364.27 - 3.0272 \times (52.0 - \text{SID Ile:Lys}), \text{ when SID Ile:Lys} < 52.0\%;$$

$$\text{ADG, g} = 364.27, \text{ if SID Lys} \geq 52.0\%$$

where the SID Ile:Lys level is expressed as a percentage.

The estimated regression equation for the BLQ model was:

$$\text{ADG, g} = 365.68 - [5.6749 \times (52.0 - \text{SID Ile:Lys})] + [0.2344 \times (52.0 - \text{SID Ile:Lys})^2], \text{ when SID Ile:Lys} < 52.0\%;$$

$$\text{ADG, g} = 365.68, \text{ if SID Ile:Lys} \geq 52.0\%$$

The estimated regression equation for the QP model was:

$$\text{ADG, g} = 98.0474 + 8.4079 \times (\text{SID Ile:Lys}) - 0.0651 \times (\text{SID Ile:Lys})^2$$

For ADFI (Fig. 2) from d 0 to 12, the BLL and QP resulted in competing fits (BIC = 603.8 and 604.4, respectively). The BLL breakpoint occurred at 50.6% SID Ile:Lys ratio [95% CI: (41.99, 59.15%)]. The QP reported maximum ADFI at 56.2% SID Ile:Lys ratio [95% CI: (48, > 65%)] with 99% of maximum intake captured at 51.6% SID Ile:Lys ratio.

The estimated regression equation for the BLL model was:

$$\text{ADFI, g} = 563.01 - 6.2844 \times (50.6 - \text{SID Ile:Lys}), \text{ when SID Ile:Lys} < 50.6\%;$$

$$\text{ADFI, g} = 563.01, \text{ if SID Ile:Lys} \geq 50.6\%$$

The estimated regression equation for the QP model was:

$$\text{ADFI, g} = -288.15 + 30.4124 \times (\text{SID Ile:Lys}) - 0.2705 \times (\text{SID Ile:Lys})^2$$

Experiment 2

From d 0 to 18 when experimental diets were fed, ADG and ADFI increased (quadratic, $P < 0.003$), but there was no evidence for differences in G:F as SID Ile:Lys ratio increased (Table 7). During the common diet period (d 18 to 32), there was no evidence for differences for ADG, but ADFI increased (linear, $P <$

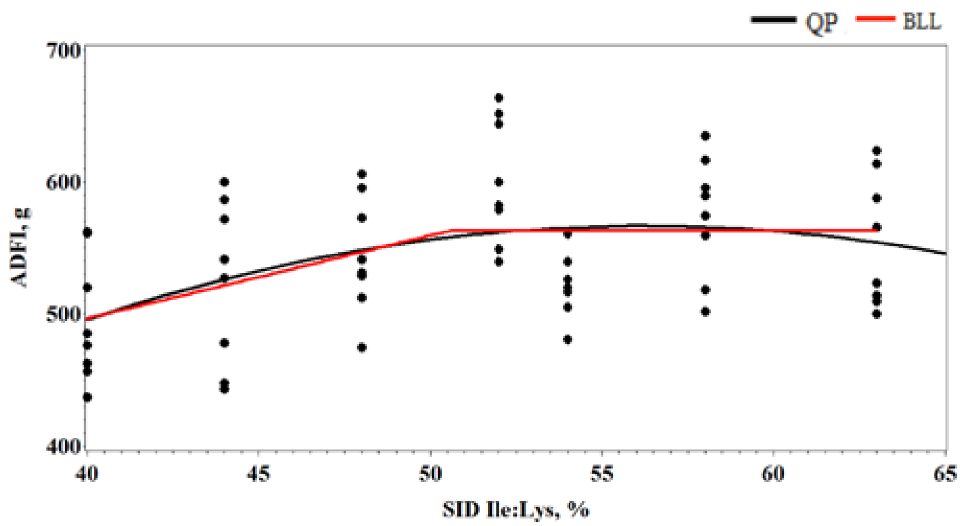

Figure 2. Estimated standardized ileal digestible (SID) Ile:Lys ratio requirement for ADFI of nursery pigs, Exp. 1. QP: BIC = 604.4; Estimated 56.2% SID Ile:Lys that maximized ADFI [95% CI: (48, > 65%)]; 99% of Max ADFI at 51.6% SID Ile:Lys. BLL: BIC = 603.8; Estimated breakpoint: 50.6% SID Ile:Lys; [95% CI:(41.99, 59.15)]. A total of 280 nursery pigs (PIC 327 × 1,050, initially 6.7 ± 0.08 kg BW) were used in a 26-d growth trial with 5 pigs per pen and 8 pens per treatment. Pigs were weaned at approximately 21 d, fed a common starter diet for 6 d post-weaning, then placed on experimental diets. Experimental diets were fed from d 0 to 12 and a common diet was fed from d 12 to 26. Quadratic polynomial (QP), broken-line linear (BLL), and broken- line quadratic (BLQ) models were fit for the experimental period to estimate SID Ile:Lys ratio to maximize ADFI. Bayesian Information Criterion (BIC) was used to determine the best fitting models; a lower value indicates a better fit to the data.

Table 7. Effects of increasing standardized ileal digestible (SID) Ile:Lys ratio on nursery pig growth performance, Exp. 2[1]

	Formulated SID Ile:Lys ratio, %[2]								Probability, $P <$	
Item	40	44	48	52	54	58	63	SEM	Linear	Quadratic
Treatment period (d 0 to 18)										
ADG, g	229	247	266	306	263	286	282	–[3]	0.001	0.016
ADFI, g	331	370	393	453	395	421	410	–[4]	0.001	0.003
G:F	0.690	0.671	0.679	0.677	0.666	0.682	0.687	0.0146	0.935	0.228
Post-treatment period (d 18 to 32)										
ADG, g	562	590	583	587	577	594	585	15.8	0.246	0.378
ADFI, g	852	906	896	940	902	928	925	25.8	0.010	0.154
G:F	0.661	0.653	0.652	0.625	0.642	0.640	0.633	0.0093	0.009	0.298
Overall (d 0 to 32)										
ADG, g	375	397	405	429	399	421	415	11.5	0.001	0.034
ADFI, g	559	605	613	666	615	643	635	18.6	0.001	0.010
G:F	0.670	0.658	0.661	0.645	0.651	0.655	0.653	0.0090	0.107	0.209
BW, kg										
d 0	6.0	6.0	6.0	6.0	6.0	6.0	6.0	0.27	0.824	0.920
d 18	10.1	10.5	10.8	11.5	10.8	11.2	11.1	0.41	0.001	0.010
d 32	18.0	18.7	19.0	19.7	18.9	19.5	19.3	0.59	0.001	0.032

[1]A total of 280 nursery pigs (DNA Genetics Line 600 × Line 241, initially 6.0 ± 0.27 kg BW) were used in a 32-d growth trial with 8 pens per treatment and 5 pigs per pen. Pigs were weaned at approximately 20 d of age. After a common diet period, pens were placed on experimental diets. Experimental diets were fed from d 0 to 18 and a common diet was fed from d 18 to 32.

[2]Low (40% SID Ile:Lys) and high (63% SID Ile:Lys) complete diets were blended upon manufacturing at the feed mill to create the 44, 48, 52, 54, and 58% SID Ile:Lys dietary treatments.

[3]Heteroskedastic modeling resulted in SEM = 11.5 for 40, 44, 52, 54, 58, and 63% dietary treatments and 3.5 for 48% SID Ile:Lys ratio treatment.

[4]Heteroskedastic modeling resulted in SEM = 17.3 for 40, 44, 52, 54, 58, and 63% dietary treatments and 7.9 for 48% SID Ile:Lys ratio dietary treatment.

0.010) and G:F decreased (linear, $P < 0.009$) for pigs previously fed diets with increasing SID Ile:Lys ratio. For the overall period (d 0 to 32), ADG and ADFI increased (quadratic, $P < 0.034$) with increasing SID Ile:Lys ratio with no differences in G:F. Finally, BW was increased (quadratic, $P < 0.032$) at the end of phase 1 and at the conclusion of the experiment with increasing SID Ile:Lys ratio.

For ADG (Fig. 3) from d 0 to 18, the BLL and QP were competing best fit models (BIC = 541.8 and

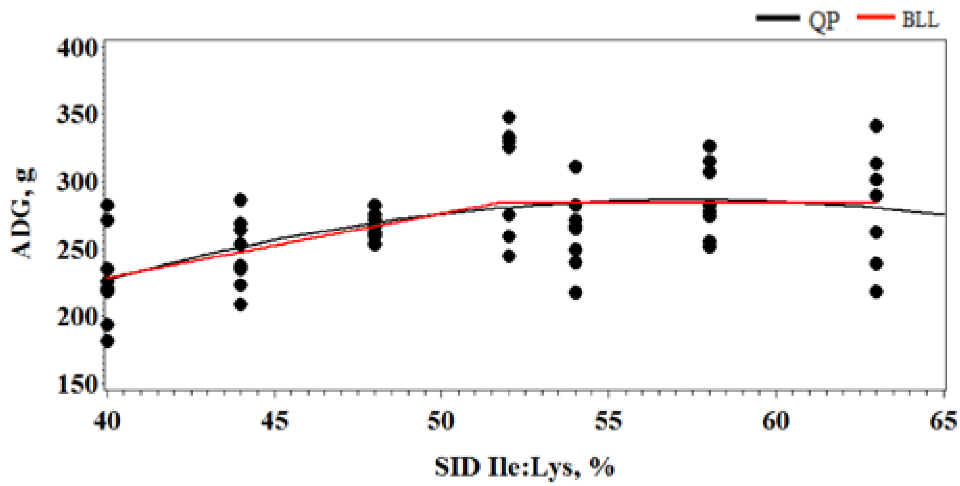

Figure 3. Estimated standardized ileal digestible (SID) Ile:Lys ratio requirement for ADG of nursery pigs, Exp. 2. QP: BIC = 543.3; Estimated 58.3% SID Ile:Lys that maximized ADG; [95% CI: (49, > 65%)]; 99% of Max ADG at 54.3% SID Ile:Lys. BLL: BIC = 541.8; Estimated breakpoint: 51.8% SID Ile:Lys; [95% CI: (47.65, 55.93)]. A total of 280 nursery pigs (DNA 600 × 241, initially 6.0 ± 0.27 kg BW) were used in a 32-d growth trial with 5 pigs per pen and 8 pens per treatment. Pigs were weaned at approximately 20 d of age. One replication was fed a common starter diet for 3 d due to increased weaning BW, and the other seven replications were fed a common starter diet for 6 d post-weaning, then placed on experimental diets. Experimental diets were fed from d 0 to 18 and a common diet was fed from d 18 to 32. Quadratic polynomial (QP), broken-line linear (BLL), and broken-line quadratic (BLQ) models were fit for the experimental period to estimate SID Ile:Lys ratio to maximize ADG. Bayesian Information Criterion (BIC) was used to determine the best fitting models; a lower value indicates a better fit to the data.

543.3, respectively). The BLL breakpoint occurred at 51.8% SID Ile:Lys ratio [95% CI: (47.65, 55.93%)]. The QP model resulted in maximum ADG at 58.3% SID Ile:Lys ratio [95% CI: (49, > 65%)] with 99% of maximum performance captured with 54.3% SID Ile:Lys ratio.

The estimated regression equation for the BLL model was:

$$\text{ADG, g} = 284.29 - 4.7304 \times (51.8 - \text{SID Ile:Lys}), \text{ when SID Ile:Lys} < 51.8\%;$$

$$\text{ADG, g} = 284.29, \text{ if SID Ile:Lys} \geq 51.8\%$$

The estimated regression equation for the QP model was:

$$\text{ADG, g} = -311.01 + 20.449 \times (\text{SID Ile:Lys}) - 0.1753 \times (\text{SID Ile:Lys})^2$$

For ADFI (Fig. 4) from d 0 to 18 modeled with heterogeneous variance, the QP and BLQ resulted in similar competing fits (BIC = 591.0 and 591.7, respectively). The BLQ breakpoint occurred at 52.0% SID Ile:Lys ratio [95% CI: (51.95, 52.05%)]. The QP reported maximum ADFI at 57.2% SID Ile:Lys ratio [95% CI: (49, > 65%)] with 99% of maximum intake captured at 53.5% SID Ile:Lys ratio.

The estimated regression equation for the BLQ model was:

$$\text{ADFI, g} = 419.44 - [6.1716 \times (52.0 - \text{SID Ile:Lys})] - [0.08475 \times (52.0 - \text{SID Ile:Lys})^2], \text{ when SID Ile:Lys} < 52.0\%;$$

$$\text{ADFI, g} = 419.44, \text{ if SID Ile:Lys} \geq 52.0\%$$

The estimated regression equation for the QP model was:

$$\text{ADFI, g} = -588.47 + 35.277 \times (\text{SID Ile:Lys}) - 0.3082 \times (\text{SID Ile:Lys})^2$$

DISCUSSION

In experiments to determine the Ile:Lys ratio required by pigs, researchers use 1 of 2 different approaches in diet formulation, either with or without SDBC. Without SDBC it is difficult to obtain diets with low enough Ile:Lys ratios to characterize the lower part of the dose response curve. Use of higher level of SDBC (ex. 7.5%) allows researchers to reach the lower part of the dose response curve. Unfortunately, SDBC also contain high level of Leu (NRC, 2012). The Ile requirement increases when other BCAA are in excess, particularly due to an antagonistic effect with excess Leu (Morales et al., 2016). This mechanism occurs when elevated Leu increases levels of the enzyme complex branched-chain keto-acid dehydrogenase, which increases degradation of all BCAA; thus catabolizing Ile and increasing the requirement

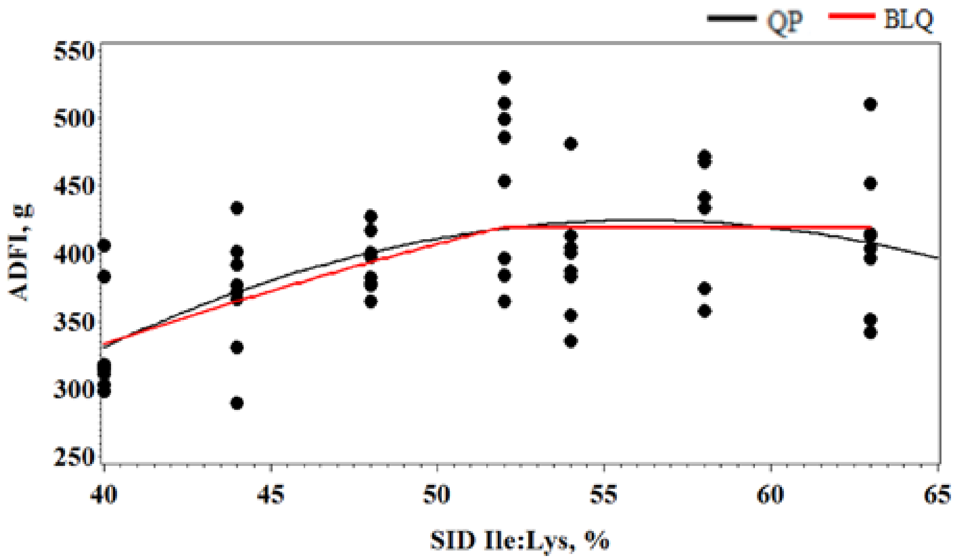

Figure 4. Estimated standardized ileal digestible (SID) Ile:Lys ratio requirement to maximize ADFI for nursery pigs, Exp. 2. QP: BIC = 591; Estimated 57.2% SID Ile:Lys that maximized ADFI; [95% CI: (49, > 65%)]; 99% of Max ADFI 53.5% SID Ile:Lys. BLQ: BIC = 591.7; Estimated breakpoint: 52.0% SID Ile:Lys; [95% CI:(51.95, 52.05)]. A total of 280 nursery pigs (DNA 600 × 241, initially 6.0 ± 0.27 kg BW) were used in a 32-d growth trial with 5 pigs per pen and 8 pens per treatment. Pigs were weaned at approximately 20 d of age. One replication was fed a common starter diet for 3 d due to in- creased weaning BW, and the other seven replications were fed a common starter diet for 6 d post-weaning, then placed on experimental diets. Experimental diets were fed from d 0 to 18 and a common diet was fed from d 18 to 32. Quadratic polynomial (QP), broken-line linear (BLL), and broken-line quadratic (BLQ) models were fit for the experimental period to estimate SID Ile:Lys ratio to maximize ADFI. Bayesian Information Criterion (BIC) was used to determine the best fitting models; a lower value indicates a better fit to the data.

(Langer et al., 2000; Wiltafsky et al., 2010). Gietzen and Magrum (2001) also describe an anorexic response as a result of BCAA imbalance or deficiency. Thus, as feed intake is critical during the post-weaning period, it is important to understand the Ile requirement in relation to the level of SDBC in the diets. We used an alternative approach to diet formulation using 1.5% SDBC and diets that incorporated 10% field peas which allowed for further reduction of SID Ile:Lys but limiting SID Leu levels at no greater than 109% of Lys. The field peas were from a known batch of peas analyzed for total AA content with SID coefficients determined by Mathai (2015). Due to the challenges associated with formulating diets with low Ile while limiting Leu, there are relatively few reports in scientific literature where this approach was used.

The Ile requirement seems to increase when SDBC are included in the diet due to antagonistic interactions previously described. Htoo et al. (2014) incorporated SDBC ranging from 3.7 to 4.1% of the diet, depending on the phase. This strategy allowed diets to be below the estimated Ile requirement, but also contain moderate Leu levels at no greater than 130% of Lys. They observed a 51 and a 54% SID Ile:Lys ratio requirement for 10- to 22-kg and 24- to 39-kg pigs, respectively, determined using averages from both curvilinear and exponential regression models. Kerr et al. (2004) evaluated the apparent ileal digestible Ile:Lys requirement for 7- to 11-kg pigs using 7.5% dietary SDBC and found the requirement was 61% Ile of Lys. Similarly, Wiltafsky et al. (2009) using 7.5% dietary blood cells (1.61% SID dietary Leu) found a requirement as high as 59% SID Ile:Lys, but the requirement was only 54% in diets that included corn gluten rather than SDBC. Also, Nørgaard et al. (2013) evaluated the SID Ile:Lys ratio requirement for pigs weighing 8- to 18-kg and found that 52% SID Ile:Lys was sufficient in diets containing no blood products, agreeing with the results from the broken-line models in this experiment. Similarly, Soumeh et al. (2014) using SDBC-free diets found that 52% SID Ile:Lys ratio was the requirement for ADG and ADFI but that the requirement for G:F was slightly lower at 48% SID Ile:Lys ratio for 8- to 15-kg pigs. Barea et al. (2009) evaluated individually-housed 11- to 23-kg pigs and determined that for diets without SDBC, no greater than 50% SID Ile:Lys was necessary. A meta-analysis by van Milgen et al. (2012) also reported that diets containing no blood products required at least 50% SID Ile:Lys (and at least 55% SID Ile:Lys in diets with blood products).

Our estimates for the SID Ile:Lys ratio required to maximize performance based on quadratic models were much higher than levels in aforementioned literature, at 64% of Lys, while the broken-line models resulted in maxima very close to the NRC (2012) requirements. However, further evaluation of the quadratic models indicated that 99% of maximum ADG or maximum ADFI can be achieved using 51 or 57 SID Ile:Lys ratios, respectively, which is similar to the 52% SID Ile:Lys estimates from the broken-line models. This indicates that even though the point minimum ratio for the broken line is much lower than the maximum point from the quadratic model, the majority of the responses with the quadratic models are achieved near the broken line point estimates. This helps explain why the 2 models had similar fit and overall have a similar interpretation once they are reviewed in context. Also, this illustrates that assumption of functional form to describe the response can be a source of variation between the results and conclusions of different studies.

In conclusion, these experiments demonstrate that the SID Ile requirement for 6- to 11-kg nursery pigs is approximately 52% of Lys for ADG and ADFI using broken line models and maximum response as high as 64% of Lys using quadratic models. These data validate that the Ile requirement for 6- to 11-kg pigs appears to be similar to NRC (2012) requirement estimates of 51.1% for the 7- to 11-kg nursery pig.

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