

Increased lysine: metabolizable energy ratio improves grower pig performance during a porcine reproductive and respiratory syndrome virus challenge

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ABSTRACT: Porcine reproductive and respiratory syndrome virus (PRRSV) reduces grower pig performance. The amino acid (AA) requirements and lysine:metabolizable energy ratio (Lys:ME) of health-challenged pigs for optimum performance are poorly understood. Two experiments were conducted to evaluate the effect of increasing standardized ileal digestible (SID) Lys:ME (g SID Lys per Mcal ME) on growth performance during a PRRSV challenge. In Exp. 1, a total of 379 barrows (51.3 ± 0.3 kg body weight [BW]) were allotted to one of six diets (1.87 to 3.41 Lys:ME) for a 35-d growth study. In Exp. 2, a total of 389 barrows (29.2 ± 0.23 kg BW) were allotted to one of six diets (2.39 to 3.91 Lys:ME) for a 49-d growth study. These isocaloric diets represented 80% to 130% of National Research Council (NRC) SID Lys requirement. For each experiment, pigs were randomly allotted across two barns of 24 pens each with seven to nine pigs per pen (four pens per diet per health status). On day 0, one barn was inoculated with live PRRSV, one barn sham inoculated (control), and all pigs were started on experimental diets. Pen growth performance and feed intake were recorded weekly and gain-to-feed ratio (G:F) was calculated. Breakpoint analysis was used to determine the Lys:ME that maximized average daily gain (ADG) and G:F over the 35 or 49-d test periods for Exp. 1 and 2, respectively. In

Exp. 1, increasing Lys:ME increased ADG (quadratic $P = 0.01$) and G:F (linear and quadratic $P = 0.04$) in control pigs over 35 d. In PRRSV-infected pigs, ADG and G:F increased linearly with increasing Lys:ME ($P < 0.01$). The Lys:ME for optimum ADG and G:F during PRRSV challenge was 2.83 and 3.17, respectively, compared to 2.24 and 2.83, respectively, in control pigs using a one-slope broken-line model. In Exp. 2, pigs in the control barn became naturally infected after 21 days post inoculation. Before infection, ADG and G:F increased with increasing Lys:ME in control and PRRSV-infected pigs (linear and quadratic $P < 0.05$), and optimum ADG and G:F were achieved at 3.02 and 2.92 Lys:ME, respectively, in PRRSV-infected pigs compared to 2.82 and 3.22 Lys:ME, respectively, in control pigs. Over the 49-d period, increasing Lys:ME improved ADG ($P < 0.01$, linear and quadratic) and G:F (linear $P < 0.01$) in naturally infected pigs. The response was similar in experimental infection for ADG ($P < 0.01$, linear and quadratic) and G:F (linear $P = 0.01$). The optimum ratio for ADG (2.86 vs. 3.12 Lys:ME) and G:F (3.18 vs. 3.08 Lys:ME) were similar between natural and experimental infection. In summary, increasing Lys:ME by 10% to 20% above NRC requirements improved performance and feed efficiency during an experimental and natural PRRSV challenge.

Key words: lysine, metabolizable energy, pig, performance, porcine reproductive and respiratory syndrome virus

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INTRODUCTION

Nutritional requirements have been well established for healthy pigs; however, requirements for pigs facing health challenges are largely unexplored, particularly amino acid (AA) requirements and these requirements in relation to energy intakes. It has been established that pig performance and lean tissue accretion rates are decreased due to different pathogens (Escobar et al., 2004; Curry et al., 2017; Schweer et al., 2017; Helm et al., 2018); however, it is not known if this is a result of decreased feed intake. In addition, this may be due to a repartitioning of nutrients, specifically AA, to meet altered metabolic and immune needs (Klasing and Calvert, 1999). Lysine is the first limiting AA for healthy pigs fed corn–soybean meal diets; however, AA pertinent to the immune system and its activation may differ from that of growth (Reeds et al., 1994; Le Floc'h et al., 2004).

Interestingly, Lys requirements (g per day basis) are reduced in immune-stimulated pigs compared to control pigs (Williams et al., 1997b, 1997c). This is due to a greater capacity for proteinaceous tissue accretion in healthy pigs as partial efficiency for Lys utilization may not be altered due to health status (Williams et al., 1997a). In addition, adequate energy is essential for a proper immune response. Diets deficient in protein and energy can lead to reduced growth during parasite (*Trichuris suis* also known as whipworm) infection (Pedersen et al., 2002).

Porcine reproductive and respiratory syndrome virus (PRRSV) is one of the most economically significant pathogens to the swine industry. However, research pertaining to this virus's impact on nutritional requirements in pigs is minimal. Our group has recently reported in growing pigs that PRRSV reduces lean tissue accretion rates (Schweer et al., 2017), but basal endogenous losses of many AA and standardized ileal digestibilities (SID) of AA are not different (Schweer et al., 2018). Therefore, the objective of these studies was to evaluate the effects of graded levels of g SID Lys per Mcal ME (lysine:metabolizable energy ratio [Lys:ME]) on pig performance during a health challenge in the growing phase. This will allow for the optimal Lys:ME to be defined for PRRSV-challenged pigs.

MATERIALS AND METHODS

All procedures adhered to the ethical and humane use of animals for research and were approved by the Iowa State University Institutional Animal

Care and Use Committee (IACUC# 8-16-8330-S). This work was conducted between January and November 2017 in Ames, IA.

Two experiments were conducted to determine the ideal SID Lys:ME for grow-finish barrows (purebred Maschhoffs proprietary line Duroc sires by commercial Yorkshire–Landrace F1 females) during a PRRSV challenge. In both experiments, pigs were split across two identical barns with identical ventilation systems and temperature set points. The two barns used were approximately 15.2 m apart, contained two rooms each, and each room had 12 large pens plus 2 spare pens. Each pen contained a double-sided feeder and two nipple drinkers. In each pen, pigs had greater than 0.9 m² per pig floor space. All test pen floor spaces, feeders, and waterers in each room were identical. One barn was maintained as a PRRS-negative control and the other inoculated with a live PRRSV isolate (open reading frame [ORF] 5 sequence 1-18-4; Schweer et al., 2017). Pigs in the PRRSV barn were inoculated on days post inoculation (dpi) 0 with 2 mL of live PRRSV (1 mL intramuscular and 1 mL intranasal; 10⁶ genomic PRRSV units per mL) whereas the control barn received a sham saline inoculation. Pigs were allowed unrestricted access to feed and water. During the challenge period, pigs were fed one of six experimental diets; body weight (BW) and feed disappearance were measured weekly to determine average daily gain (ADG), average daily feed intake (ADFI), and to calculate feed efficiency. Experimental diets (Tables 1 and 2) were corn–soybean meal based and were formulated to be isocaloric (ME basis) and meet or exceed the nutritional requirements of 50 to 100 kg and 25 to 50 kg pigs in Exp. 1 and 2, respectively (NRC, 2012). There was a stepwise increase in SID Lys:ME and minimum ratios of SID Thr, Trp, Met, Ile, and Val to SID Lys were maintained. The dietary SID Lys:ME levels were achieved by increasing soybean meal and L-Lys-hydrochloride (HCl). By design, as Lys:ME increased so did crude protein (CP), but the essential AA to Lys ratios were maintained using crystalline AA. The diet was formulated so a majority of the SID Lys requirement was met with soybean meal to maintain commercial relevance. These diets correlated to 80%, 90%, 100%, 110%, 120%, and 130% of NRC (2012) Lys requirement that has been previously validated internally within the Maschhoffs production system for 50 to 100 kg and 25 to 50 kg pigs used in Exp. 1 and 2, respectively.

Table 1. Experiment 1 diet composition, as fed basis

Ingredients, %	SID Lys:ME, g/Mcal					
	1.87	2.18	2.49	2.80	3.11	3.41
Corn	87.16	84.13	81.07	77.74	73.95	70.29
Soybean meal, 48%	9.75	12.75	15.74	19.10	22.92	26.61
Limestone	1.00	1.01	1.02	1.02	1.02	1.03
Monocalcium phosphate, 21%	1.13	1.07	1.05	0.94	0.86	0.79
Salt	0.51	0.51	0.51	0.51	0.51	0.51
L-Lysine·HCl	0.27	0.31	0.34	0.37	0.38	0.39
Commercial VTM ¹	0.11	0.11	0.11	0.11	0.11	0.11
L-Threonine	0.05	0.06	0.08	0.10	0.10	0.11
DL-Methionine	0.02	0.05	0.08	0.11	0.13	0.15
Optiphos 1000	—	—	—	0.004	0.006	0.009
Calculated composition						
DM, %	85.6	85.7	85.8	85.9	86.0	86.1
CP, %	11.4	12.6	13.9	15.2	16.8	18.3
ME, Mcal/kg	3.28	3.27	3.27	3.27	3.27	3.26
Total calcium, %	0.65	0.65	0.66	0.65	0.65	0.65
STTD phosphorus, %	0.32	0.32	0.32	0.31	0.30	0.31
SID AA						
Lys	0.61	0.71	0.81	0.92	1.02	1.11
Thr:Lys	0.60	0.60	0.60	0.60	0.60	0.60
Met:Lys	0.30	0.32	0.33	0.34	0.35	0.35
Met+Cys:Lys	0.57	0.57	0.57	0.57	0.57	0.57
Trp:Lys	0.16	0.16	0.16	0.16	0.17	0.17
Ile:Lys	0.58	0.58	0.57	0.57	0.58	0.59
Val:Lys	0.71	0.68	0.66	0.65	0.65	0.65
SID Lys:ME, g/Mcal	1.87	2.18	2.49	2.80	3.11	3.41
Total Lys, %	0.70	0.80	0.91	1.02	1.13	1.23
Analyzed composition						
DM, %	85.8	85.8	85.9	87.3	87.1	87.0
GE (Mcal)	3.75	3.82	3.80	3.86	3.87	3.88
CP, %	13.6	15.3	16.3	18.4	20.3	22.8
Total AA:Lys						
Thr:Lys	0.61	0.63	0.61	0.62	0.67	0.59
Met:Lys	0.28	0.29	0.29	0.31	0.33	0.28
Met+Cys:Lys	0.58	0.58	0.55	0.57	0.59	0.50
Trp:Lys	0.15	0.16	0.16	0.16	0.19	0.17
Ile:Lys	0.57	0.56	0.64	0.65	0.68	0.56
Val:Lys	0.71	0.68	0.72	0.72	0.74	0.63
Lys, total %	0.76	0.87	0.96	1.04	1.14	1.27

¹VTM = Vitamin-trace mineral premix, which supplied per kilogram of diet: vitamin A, 8,820 IU; vitamin D₃, 1,653 IU; vitamin E, 33.1 IU; vitamin K, 4.4 mg; riboflavin, 6.6 mg; niacin, 38.9 mg; pantothenic acid, 22.1 mg; vitamin B₁₂, 0.04 mg; I, 1.1 mg as potassium iodide; Se, 0.30 mg sodium selenite; Zn, 60.6 mg as zinc oxide; Fe, 36.4 mg as ferrous sulfate; Mn, 12.1 mg as manganous oxide; and Cu, 3.6 mg as copper sulfate. STTD = standardized total tract digestible.

Experiment 1, 50 to 100 kg BW pigs

In Exp. 1, a total of 379 barrows (51.3 ± 0.32 kg BW) were randomly allotted to one of six dietary treatments with four pens per treatment per health status and 7 to 8 pigs per pen. Before arrival, all pigs were vaccinated for *Mycoplasma hyopneumoniae*, porcine circovirus, erysipelas, and ileitis, and were serologically negative for PRRSV as determined by polymerase chain reaction (PCR). Pigs were given

a 14-d acclimation period during which all pigs were fed a common diet. At the time of PRRSV inoculation, pigs were started on experimental diets (Table 1), and performance was measured for 35 d. Diets were formulated to contain 1.87, 2.18, 2.49, 2.80, 3.11, and 3.41 SID Lys:ME, representing 80%, 90%, 100%, 110%, 120%, and 130% of National Research Council (NRC) requirement, respectively. Weekly after PRRSV inoculation, the

Table 2. Experiment 2 diet composition, as fed basis

Ingredients, %	SID Lys:ME, g/Mcal					
	2.33	2.63	2.94	3.24	3.55	3.85
Corn	82.16	79.11	75.59	71.87	68.02	64.29
Soybean meal, 48% CP	14.55	17.52	21.08	24.84	28.73	32.49
Limestone	0.96	0.98	0.98	0.99	0.99	1.00
Monocalcium phosphate, 21%	1.01	0.99	0.89	0.80	0.72	0.64
Salt	0.51	0.51	0.51	0.51	0.51	0.51
L-Lysine-HCl	0.32	0.35	0.37	0.38	0.39	0.40
Beef tallow	0.25	0.25	0.25	0.25	0.25	0.25
Commercial VTM ¹	0.11	0.11	0.11	0.11	0.11	0.11
L-Threonine	0.07	0.09	0.10	0.10	0.11	0.12
DL-Methionine	0.06	0.09	0.12	0.14	0.16	0.18
Optiphos 1000	0.00	0.00	0.008	0.008	0.009	0.012
Calculated composition						
DM, %	86.2	86.3	86.3	86.4	86.5	86.6
CP, %	13.0	14.2	15.6	17.2	18.7	20.2
ME, Mcal/kg	3.29	3.29	3.29	3.29	3.29	3.29
Total calcium, %	0.63	0.64	0.63	0.63	0.63	0.63
STTD phosphorus, %	0.31	0.31	0.30	0.30	0.29	0.28
SID AA						
Lys	0.77	0.86	0.97	1.07	1.17	1.27
Thr:Lys	0.60	0.60	0.60	0.60	0.60	0.60
Met:Lys	0.32	0.33	0.34	0.34	0.35	0.35
Met+Cys:Lys	0.57	0.57	0.57	0.57	0.57	0.57
Trp:Lys	0.16	0.16	0.16	0.17	0.17	0.17
Ile:Lys	0.58	0.58	0.58	0.59	0.59	0.60
Val:Lys	0.67	0.66	0.65	0.65	0.65	0.65
SID Lys:ME, g/Mcal	2.33	2.63	2.94	3.24	3.55	3.85
Lys, total %	0.86	0.97	1.08	1.18	1.29	1.40
Analyzed composition						
DM, %	86.3	86.1	86.5	86.6	86.5	86.8
GE, Mcal/kg	3.73	3.80	3.81	3.84	3.86	3.84
CP, %	14.1	15.7	16.1	17.9	20.2	20.8
Total AA:Lys						
Thr:Lys	0.57	0.61	0.64	0.61	0.62	0.61
Met:Lys	0.26	0.31	0.32	0.32	0.30	0.30
Met+Cys:Lys	0.49	0.57	0.56	0.55	0.53	0.53
Trp:Lys	0.15	0.18	0.18	0.17	0.18	0.17
Ile:Lys	0.53	0.64	0.64	0.59	0.61	0.62
Val:Lys	0.63	0.71	0.70	0.64	0.66	0.66
Lys, total %	1.00	1.00	1.07	1.21	1.38	1.44

¹VTM=Vitamin-trace mineral premix, which supplied per kilogram of diet: vitamin A, 8,820 IU; vitamin D₃, 1,653 IU; vitamin E, 33.1 IU; vitamin K, 4.4 mg; riboflavin, 6.6 mg; niacin, 38.9 mg; pantothenic acid, 22.1 mg; vitamin B₁₂, 0.04 mg; I, 1.1 mg as potassium iodide; Se, 0.30 mg sodium selenite; Zn, 60.6 mg as zinc oxide; Fe, 36.4 mg as ferrous sulfate; Mn, 12.1 mg as manganous oxide; and Cu, 3.6 mg as copper sulfate. STTD = standardized total tract digestible.

same two pigs per pen were bled for PRRSV PCR and enzyme-linked immunosorbent assay (ELISA). After the experimental period, all pigs were fed a common multiphase diet that met or exceeded [NRC \(2012\)](#) requirements until pigs reached market BW (approximately 128 kg BW), at which time pigs were slaughtered, and carcass data collected from the slaughter plant (JBS, Marshalltown, IA). Pigs were shipped for slaughter in two groups approximately 5 d apart and these two groups were based

on BW. Shipping and pre-slaughter handling were the same for control and PRRSV-infected pigs.

Experiment 2, 25 to 50 kg BW pigs

In Exp. 2, a total of 389 barrows (29.2 ± 0.23 kg BW) were vaccinated for *M. hyopneumoniae*, porcine circovirus, erysipelas, and ileitis before arrival, and serologically negative for PRRSV as determined by PCR. Barrows were randomly allotted to

one of six dietary treatments formulated to contain 2.33, 2.63, 2.94, 3.24, 3.55, and 3.85 SID Lys:ME, representing 80%, 90%, 100%, 110%, 120%, and 130% of NRC requirement, respectively (Table 2). Each treatment had four pens per treatment per health status with seven to nine pigs per pen. After a 10-d acclimation on a common diet, pigs were inoculated with PRRSV and started on experimental diets for a 49-d growth study. Between 21 and 28 dpi, the control barn became naturally infected with PRRSV and were confirmed positive by serum PCR. The PRRSV strain isolated from the control barn was considered identical to the challenge isolate used in the PRRS barn by ORF-5 sequence. After the experimental period, all pigs were fed a common multiphase diet that met or exceeded NRC (2012) requirements until pigs reached market BW (approximately 130 kg BW). In Exp. 2, carcass data were not obtained from the slaughter plant.

Diet Analysis

Analysis of gross energy (GE) in experimental diets in both experiments was carried out using bomb calorimetry (Oxygen Bomb Calorimeter 6200; Parr Instrument Company, Moline, IL). Dietary dry matter (DM) in both experiments was carried out in a commercial laboratory (Midwest Labs, Omaha, NE). Dietary AA and nitrogen (N) analysis were conducted by Ajinomoto Heartland, Inc (Eddyville, IA). AA and N analysis were performed using method 994.12, 999.13, and 990.03 according to AOAC (2007) methods, and CP was calculated ($N \times 6.25$).

Blood Collection and Analysis

In both experiments, 8 to 10 mL blood samples were collected from the jugular vein into serum tubes (BD Vacutainer, Franklin Lakes, NJ) while pigs were snare restrained. In Exp. 1, the same two pigs per pen were bled weekly during the 35-d challenge period. In Exp. 2, six pigs per room (12 pigs per barn) were randomly selected and bled weekly during the 49-d challenge period. Serum from these pigs was pooled within room after centrifugation. Serum was allowed to clot then separated by centrifugation ($2,000 \times g$, 15 min at 4 °C), aliquoted and submitted to the Iowa State University Veterinary Diagnostic Laboratory. Real-time reverse transcription PCR (RT-PCR) and serum antibody testing for PRRSV was performed using commercial reagents (VetMAX PRRSV NA and EU reagents; Thermo Fisher Scientific, Waltham,

MA) and a commercial ELISA kit (HerdCheck PRRS X3; IDEXX Laboratories, Inc, Westbrook, ME), respectively.

Statistics

Data within health status were analyzed using the PROC MIXED procedure of SAS (SAS Institute Inc, Cary, NC) for linear and quadratic effects of increasing SID Lys:ME. Pen served as the experimental unit in both experiments. Data were considered significant if $P \leq 0.05$ and a trend if $P \leq 0.10$. For all experiments, both one-slope straight broken-line and quadratic broken-line analysis was conducted as described by Robbins et al. (2006) to estimate SID Lys:ME requirement for ADG and G:F of each health status (control or PRRS pigs). In Exp. 1, breakpoint analysis was performed on performance over the 35-d experimental period. In Exp. 2, breakpoint analysis was performed on performance from 0 to 21 dpi, the period for which control pigs were negative for PRRSV. Further, breakpoint analysis was performed over the 49-d experimental period (Exp. 2) to determine if SID Lys:ME requirements are similar between pigs naturally and experimentally infected with PRRSV.

RESULTS

Diet Analysis

Experimental diets were formulated to contain 1.87, 2.18, 2.49, 2.80, 3.11, and 3.41 and 2.33, 2.63, 2.94, 3.24, 3.55, and 3.85 g SID Lys per Mcal ME in Exp. 1 and 2, respectively (Tables 1 and 2, respectively). Proximate and AA analysis of the diets determined that experimental diets were formulated similarly to the calculated values. The ratio of SID Thr, Met + Cys, Trp, Ile, and Val to SID Lys remained the same across all diets. As expected, CP increased as soybean meal inclusion increased.

Breakpoint Analysis

To determine the optimal dietary Lys:ME for control and PRRS-infected pigs, we analyzed and reported data using both straight broken-line and quadratic broken-line models. Nonlinear regression analysis showed that both models fit the data similarly (based on the R^2 values). We reported two models as selection of a specific model can dramatically affect the identification of the requirement (Robbins et al., 2006; Nemechek et al., 2011). Also noted by Robbins et al. (2006), in a curvilinear response the

intersection of a straight broken-line and quadratic regression curve may more accurately define a requirement that can alternatively be defined using a quadratic broken-line model. This multiple-model approach has been reported in evaluating lysine requirements of nursery pigs (Jones et al., 2014).

Experiment 1

In Exp. 1, there were two mortalities in the control barn and three in the PRRS barn. Both pigs in the control barn succumbed to hemorrhagic bowel syndrome. Two unthrifty pigs in the PRRS barn were euthanized shortly after arrival, and the third was determined to expire from porcine dermatitis and nephropathy syndrome. There were no associations between mortality and dietary treatment.

Before experimental inoculation with PRRSV in both experiments, all pigs were negative for PRRSV and antibody. In Exp. 1, control pigs remained negative for PRRSV and antibody throughout the 35-d experimental diet period and to market, as expected. No diet or diet × dpi interactions were detected for PRRSV PCR Ct value or Log₁₀ PRRSV genomic content (Table 3). Similarly, no differences were detected for PRRSV antibody ($P > 0.10$). Expectedly, PRRSV Ct value and Log₁₀ genomic content decreased over time whereas

PRRSV antibody increased causing a main effect of dpi ($P < 0.001$).

Before feeding experimental diets and inoculation, growth performance and feed efficiency were not different in control or PRRSV-infected pigs (Table 4). Over the 35-d test period, control pig ADG increased as SID Lys:ME increased (quadratic, $P = 0.013$). G:F increased (linear, $P = 0.039$; quadratic, $P = 0.037$) as SID Lys:ME increased. Feed intake was not different over the 35-d test period in control pigs. In the post-challenge period, when all pigs were on a common diet, there were no performance differences ($P > 0.10$, data not shown). Pig growth and feed intake from 0 dpi to market (76-d period) were not different ($P > 0.10$); however, G:F increased up to 3.11 SID Lys:ME resulting in a significant quadratic effect ($P = 0.040$). Over the 35-d period, PRRSV-infected pig ADG and G:F increased linearly with increasing SID Lys:ME ($P = 0.001$ and $P = 0.002$, respectively), and ADFI tended to increase (linear, $P = 0.068$). Similar to control pigs, there was no difference after 35 dpi when all pigs were on a common diet (data not shown). From inoculation to market (78 ± 2 d), ADG increased linearly with increasing SID Lys:ME ($P = 0.011$); however, ADFI and G:F were not different ($P > 0.10$).

Table 3. Effect of increasing SID Lys:ME on PRRS viremia and antibody, Exp. 1

Parameter ¹	SID Lys:ME, g/Mcal						SEM	P value ²		
	1.87	2.18	2.49	2.80	3.11	3.41		Diet	dpi	Diet × dpi
PRRSV Ct value ³										
dpi 7	21.9	21.5	23.5	23.0	21.9	22.0	1.28	0.124	<0.001	0.951
dpi 14	32.8	27.3	30.0	31.4	28.8	32.3				
dpi 21	33.7	32.7	33.7	33.9	32.7	33.3				
dpi 28	37.0	34.2	37.0	36.8	36.6	36.5				
dpi 35	37.0	35.5	35.5	37.0	36.2	37.0				
Genomic PRRSV/mL ⁴										
dpi 7	7.33	7.30	6.99	7.01	7.34	7.31	0.71	0.407	<0.001	0.946
dpi 14	3.36	4.92	4.92	4.49	4.60	4.24				
dpi 21	2.40	3.41	3.11	3.76	4.12	3.93				
dpi 28	0.00	2.26	0.00	0.78	0.82	0.88				
dpi 35	0.00	1.15	1.00	0.00	0.96	0.00				
PRRSV S/P ratio ⁵										
dpi 7	0.07	0.15	0.04	0.04	0.09	0.04	0.12	0.929	<0.001	0.676
dpi 14	2.22	2.12	2.24	1.90	2.08	2.10				
dpi 21	2.28	2.29	2.30	2.13	2.18	2.24				
dpi 28	2.02	2.24	2.25	2.21	2.13	2.19				
dpi 35	2.19	2.25	2.19	2.13	2.07	2.10				

¹ $n = 4$ pens per diet, 7–8 pigs per pen.

²Main effect of diet, day post inoculation (dpi) and diet × dpi interaction.

³Cycle threshold (Ct), Ct ≥ 37.0 denotes PRRS negative.

⁴Log₁₀ transformation of PRRSV genomic content/mL.

⁵PRRS X3 antibody sample to positive (S/P) ratio, <0.40 denotes PRRS negative.

Table 4. Effect of increasing SID Lys:ME on growth performance in healthy and PRRSV-infected growing pigs, Exp. 1

Parameter ¹	SID Lys:ME, g/Mcal						SEM	P value ²	
	1.87	2.18	2.49	2.80	3.11	3.41		Linear	Quadratic
Pre-challenge ³									
Control									
Start BW, kg	36.4	36.4	36.4	36.3	36.4	36.4	0.80	0.962	0.989
ADG, kg	1.02	0.94	0.97	1.01	0.93	1.00	0.03	0.832	0.383
ADFI, kg	1.82	1.79	1.81	1.78	1.75	1.87	0.05	0.893	0.260
G:F	0.558	0.525	0.535	0.570	0.533	0.540	0.013	0.784	0.933
PRRSV									
Start BW, kg	36.8	36.7	37.0	37.0	36.9	37.0	0.76	0.811	0.928
ADG, kg	1.03	0.99	1.05	1.08	1.08	1.01	0.03	0.372	0.148
ADFI, kg	1.88	1.71	1.86	1.88	1.88	1.86	0.04	0.278	0.669
G:F	0.548	0.578	0.565	0.575	0.570	0.548	0.015	0.922	0.110
Challenge ⁴									
Control									
Start BW, kg	50.6	49.6	50.0	50.5	49.4	50.4	1.09	0.914	0.675
ADG, kg	1.05	1.11	1.14	1.12	1.13	1.11	0.02	0.069	0.013
ADFI, kg	2.79	2.91	2.85	2.83	2.71	2.87	0.06	0.695	0.891
G:F	0.375	0.383	0.403	0.395	0.418	0.388	0.009	0.039	0.037
PRRSV									
Start BW, kg	52.2	51.6	52.7	53.3	52.9	52.2	0.89	0.563	0.454
ADG, kg	0.70	0.74	0.76	0.86	0.84	0.86	0.04	0.001	0.396
ADFI, kg	2.05	1.99	2.16	2.13	2.12	2.13	0.05	0.068	0.374
G:F	0.343	0.370	0.353	0.408	0.395	0.403	0.014	0.002	0.536
Overall ⁵									
Control									
End BW, kg	128.0	130.2	131.5	130.9	130.4	130.4	2.04	0.481	0.336
ADG, kg	1.02	1.06	1.07	1.06	1.07	1.05	0.02	0.387	0.129
ADFI, kg	2.95	3.08	3.00	2.98	2.91	3.04	0.05	0.849	0.955
G:F	0.345	0.343	0.358	0.355	0.368	0.345	0.005	0.128	0.040
PRRSV									
End BW, kg	128.4	129.4	128.5	129.0	128.6	129.7	0.93	0.569	0.788
ADG, kg	0.95	0.97	0.98	0.98	0.99	1.02	0.02	0.011	0.841
ADFI, kg	2.40	2.34	2.46	2.45	2.42	2.47	0.06	0.240	0.964
G:F	0.398	0.415	0.398	0.398	0.410	0.413	0.010	0.500	0.643

¹*n* = 4 pens per diet, 7 to 8 pigs per pen.

²Linear and quadratic orthogonal contrast.

³Pre-challenge adaptation period (–14 to 0 dpi), all pigs on common diet.

⁴Challenge period (0 to 35 dpi), pigs fed experimental diets.

⁵Overall challenge period (0 dpi to market; control = 76 d, PRRS = 78 ± 2 d).

Breakpoint analysis was used to determine the optimal SID Lys:ME to maximize growth and feed efficiency in control and PRRSV-infected pigs (Figure 1). It was determined that optimal ADG in control pigs was achieved at 2.24 and 2.38 SID Lys:ME using a one-slope and quadratic broken-line model, respectively. Optimal G:F was achieved at 2.83 and 2.95 Lys:ME in a one-slope and quadratic broken-line model, respectively. In PRRSV-infected pigs, optimal ADG and G:F were achieved at 2.83 and 3.17 SID Lys:ME, respectively, using a one-slope broken-line model. When using a quadratic broken-line model, the optimal

ADG and G:F were predicted to be 3.71 and 4.22 SID Lys:ME, respectively; however, these values are outside of the maximum SID 3.41 Lys:ME diet tested and should be studied further.

Carcass composition was evaluated when pigs reached approximately 128 kg BW (Table 5). All control pigs were marketed at 76 dpi, and there was no difference in final BW (*P* > 0.10). There was a quadratic effect (*P* = 0.016) of SID Lys:ME on fat depth where fat depth decreased from 1.87 to 2.80 SID Lys:ME and increased from 2.80 to 3.41 SID Lys:ME. Concurrently, there was a linear tendency (*P* = 0.060) for lean percentage to increase

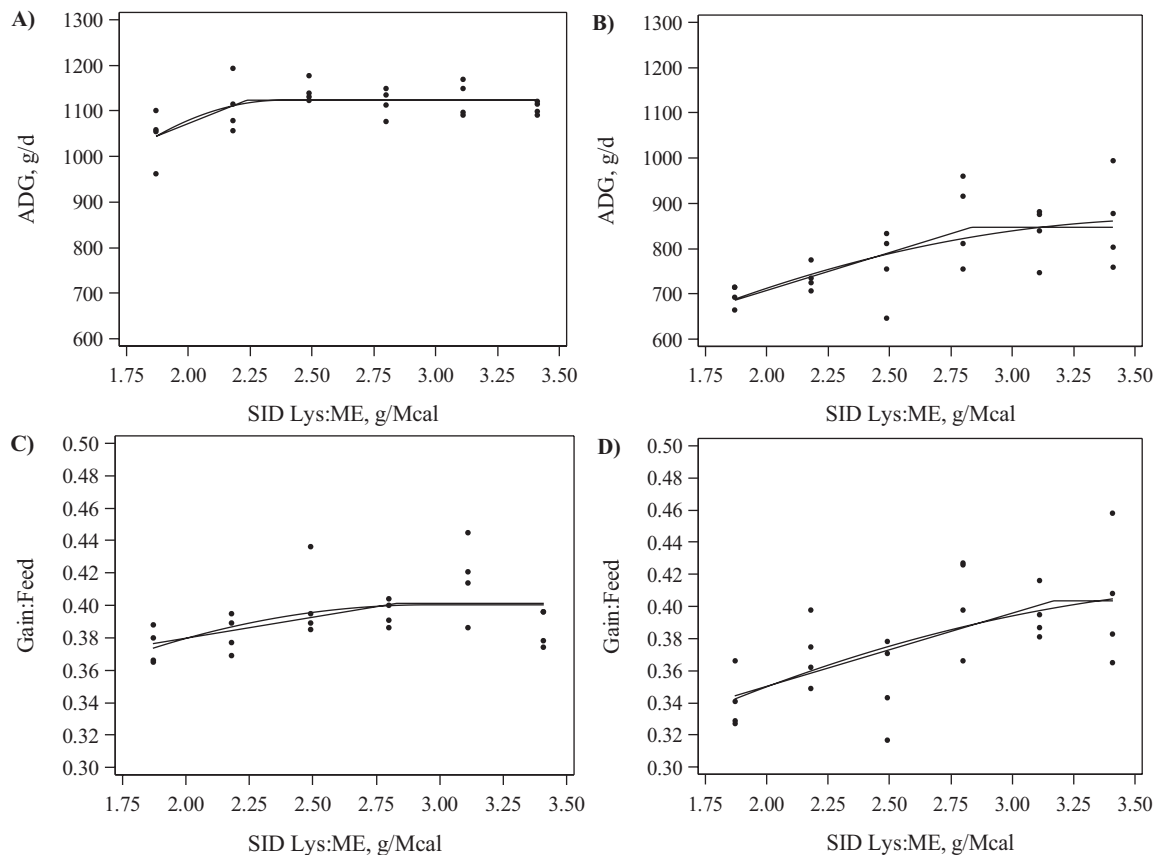


Figure 1. Data points represent treatments means from four pens per experimental diet per health status (Exp. 1). One-slope straight broken-line and quadratic broken-lines were fitted for maximum ADG (A and B) and G:F (C and D) expressed as a function of SID Lys:ME (g SID Lys per Mcal ME) over a 35-d growth period in control (A,C) and PRRSV (B,D) infected pigs, respectively. (A) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 2.24 g/Mcal (Y plateau = 1123.6 ADG; slope below requirement = -216.1 ; $r^2 = 0.38$). The quadratic broken-line model resulted in a SID Lys:ME requirement of 2.38 g/Mcal ($Y = 1123.6 - 302.5(2.38 - g \text{ SID Lys/Mcal})^2$; $r^2 = 0.38$). (B) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 2.83 g/Mcal (Y plateau = 847.4 ADG; slope below requirement = -167.2 ; $r^2 = 0.47$). The quadratic broken-line model yielded a SID Lys:ME requirement of 3.71 g/Mcal ($Y = 865.9 - 52.3(3.71 - g \text{ SID Lys/Mcal})^2$; $r^2 = 0.45$); however, this predicted requirement is outside the range of the diets tested. (C) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 2.83 g/Mcal (Y plateau = 0.401 G:F; slope below requirement = -0.026 ; $r^2 = 0.23$). The quadratic broken-line model yielded a SID Lys:ME requirement of 2.95 g/Mcal ($Y = 0.400 - 0.023(2.95 - g \text{ SID Lys/Mcal})^2$; $r^2 = 0.25$). (D) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 3.17 g/Mcal (Y plateau = 0.404 G:F; slope below requirement = -0.046 ; $r^2 = 0.41$). The quadratic broken-line model projected a SID Lys:ME requirement of 4.22 g/Mcal ($Y = 0.413 - 0.013(4.22 - g \text{ SID Lys/Mcal})^2$; $r^2 = 0.41$); however, the predicted requirement is outside the range of the experimental diets.

as SID Lys:ME increased. Hot carcass weight and dress percentage were not impacted by increasing SID Lys:ME in control pigs. In PRRSV-infected pigs, fat depth increased linearly ($P = 0.045$), and loin depth showed a strong tendency ($P = 0.059$) to decrease with increasing SID Lys:ME. Days to market decreased from 80 to 77 d as SID Lys:ME increased (linear, $P = 0.004$).

Experiment 2

The control and PRRS barn experienced three and nine mortalities, respectively. Two mortalities in the control barn were a result of porcine dermatitis and nephropathy syndrome and one from hemorrhagic bowel syndrome. In the PRRS barn, five mortalities were a result of secondary respiratory

infection, two due to gastric ulcers, one to rectal prolapse, and one to bacterial endocarditis. In both barns, there were no treatment effects on mortality. Pigs responded more severely to PRRSV infection than anticipated, therefore to decrease the impact of opportunist bacteria and avoid a high number of mortalities, antibiotics (Chlortetracycline; PharmGate Animal Health, Omaha, NE) were delivered through the water for the entire barn from 11 to 14 dpi.

As there were no differences in PRRS viremia or antibody attributed to diet in Exp. 1, pigs in Exp. 2 were randomly bled across diets to confirm PRRSV infection status in control pigs (Table 6). In Exp. 2, control pigs remained PRRSV negative until 21 dpi; however, after 21 dpi, the control pigs were naturally infected with the same PRRSV isolate used for

Table 5. Effect of increasing SID Lys:ME on carcass characteristics in control and PRRSV-infected pigs, Exp. 1

Parameter ¹	SID Lys:ME, g/Mcal						SEM	P value ²	
	1.87	2.18	2.49	2.80	3.11	3.41		Linear	Quadratic
Control									
Live weight, kg	128.0	130.2	131.5	130.9	130.4	130.4	2.04	0.481	0.336
HCW ³ , kg	97.2	100.4	99.9	97.0	98.7	97.9	1.41	0.722	0.405
Dress %	76.1	77.2	76.0	74.1	75.7	75.1	1.28	0.312	0.837
Lean %	53.4	52.0	54.1	54.9	54.2	53.7	0.79	0.205	0.355
Fat thickness, mm	20.76	20.36	20.13	18.38	20.12	20.98	0.58	0.782	0.016
Loin depth, mm	60.19	61.05	62.96	60.31	63.60	63.35	1.24	0.060	0.994
Days to market ⁴	76	76	76	76	76	76	—	—	—
PRRSV									
Live weight, kg	128.4	129.3	128.5	129.0	128.6	129.6	0.93	0.581	0.800
HCW ³ , kg	97.4	94.7	98.0	96.5	97.7	96.8	1.72	0.755	0.975
Dress %	75.8	73.2	76.2	74.8	75.9	74.7	1.27	0.920	0.918
Lean %	52.5	54.9	54.0	53.7	53.1	53.3	0.96	0.867	0.272
Fat depth, mm	19.71	18.96	20.43	20.57	21.28	21.09	0.78	0.045	0.968
Loin depth, mm	64.70	63.48	63.09	61.91	60.46	60.93	1.72	0.059	0.791
Days to market ⁴	80	80	78	78	77	77	0.88	0.004	0.627

¹n = 4 pen per diet, 7 to 8 pigs per pen.

²Linear and quadratic orthogonal contrasts.

³HCW = hot carcass weight.

⁴All pigs marketed at 76 to 80 d after start of experimental diets and were fed a common control diet.

Table 6. PRRS viremia and antibody of control and PRRSV-infected pigs, Exp. 2

Parameter ¹	Control	PRRSV	SEM	P value ²		
				Diet	dpi	Diet × dpi
PRRSV Ct value³						
dpi 7	37.0	21.3	1.55	0.115	0.010	<0.001
dpi 14	37.0	28.6				
dpi 21	35.8	34.7				
dpi 28	24.1	36.2				
dpi 49	32.2	37.0				
PRRSV S/P ratio⁴						
dpi 7	0.00	0.87	0.18	<0.001	<0.001	0.005
dpi 14	0.00	1.54				
dpi 21	0.01	1.62				
dpi 28	0.19	1.74				
dpi 49	1.78	1.75				

¹n = 4 pens per diet, 7 to 8 pigs per pen.

²Main effect of diet, day post inoculation (dpi) and diet × dpi interaction.

³Cycle threshold (Ct), Ct ≥ 37.0 denotes PRRS negative.

⁴PRRS X3 antibody sample to positive (S/P) ratio, <0.40 denotes PRRS negative.

experimental infection (ORF 5 sequence 1-18-4). The control pigs became infected with PRRSV around 21 dpi, therefore, data were analyzed as two separate challenge periods. The first challenge period, 0 to 21 dpi, represents when control pigs were not infected with PRRSV. The second period, 0 to 49 dpi, is to

determine the impact of a natural PRRSV infection compared to experimental infection.

Before experimental infection at 0 dpi, control pig performance and feed efficiency were not different (Table 7). During the first challenge period (0 to 21 dpi) when control pigs were uninfected, ADG (linear $P < 0.001$, quadratic $P = 0.020$) and G:F (linear $P < 0.001$) increased as Lys:ME increased. Feed intake increased from 2.33 to 3.24 Lys:ME and then decreased, resulting in a quadratic effect ($P = 0.039$). When breakpoint analysis was performed on 0 to 21 dpi performance, optimal ADG and G:F was achieved at 2.82 and 3.22 Lys:ME, respectively, in a one-slope broken-line model (Figure 2). In a quadratic broken-line model, optimal ADG was attained at 3.32 Lys:ME. Optimal G:F was predicted at 4.22 Lys:ME; however, this was outside the range of the experimental diets tested. In PRRSV pigs, 21 d ADG, ADFI, and G:F increased linearly with increasing Lys:ME ($P \leq 0.001$, all parameters), and ADG and G:F also demonstrated a quadratic effect ($P = 0.043$ and $P = 0.006$, respectively). Breakpoint analysis determined optimal ADG and G:F at 3.02 and 2.92, respectively, in a one-slope broken-line model and 3.41 and 3.22, respectively, in a quadratic broken-line model.

In Exp. 2, control pigs became infected with PRRSV after 21 dpi. Therefore, performance

Table 7. Effect of increasing SID Lys:ME on growth performance in healthy and PRRSV-infected pigs and natural and experimental PRRSV infection, Exp. 2

Parameter ¹	SID Lys:ME, g/Mcal						SEM	P value ²	
	2.33	2.63	2.94	3.24	3.55	3.85		Linear	Quadratic
Pre-challenge ³									
Control									
Start BW, kg	23.0	23.1	23.1	22.9	22.8	22.4	0.46	0.299	0.396
ADG, kg	0.48	0.58	0.52	0.46	0.52	0.48	0.04	0.521	0.631
ADFI, kg	1.24	1.36	1.29	1.26	1.33	1.25	0.04	0.886	0.260
G:F	0.388	0.420	0.398	0.363	0.393	0.385	0.026	0.564	0.968
PRRSV									
Start BW, kg	23.3	23.5	23.7	23.9	23.5	22.9	0.57	0.684	0.248
ADG, kg	0.59	0.53	0.58	0.66	0.64	0.62	0.02	0.008	0.808
ADFI, kg	1.43	1.30	1.35	1.41	1.35	1.31	0.04	0.264	0.943
G:F	0.408	0.405	0.435	0.463	0.473	0.473	0.014	<0.001	0.600
Challenge 1 ⁴									
Control									
Start BW, kg	28.2	29.5	28.8	27.9	28.6	27.7	0.71	0.283	0.405
ADG, kg	0.65	0.81	0.87	0.92	0.92	0.92	0.05	<0.001	0.020
ADFI, kg	1.57	1.87	1.76	1.86	1.74	1.72	0.08	0.489	0.039
G:F	0.415	0.433	0.493	0.500	0.533	0.535	0.012	<0.001	0.123
PRRSV									
Start BW, kg	29.7	29.1	30.0	31.1	30.5	29.7	0.73	0.413	0.323
ADG, kg	0.19	0.26	0.43	0.45	0.39	0.49	0.04	<0.001	0.043
ADFI, kg	0.71	0.74	0.90	0.96	0.88	0.98	0.06	0.001	0.262
G:F	0.265	0.345	0.478	0.470	0.443	0.493	0.028	<0.001	0.006
Challenge 2 ⁵									
Natural infection									
ADG, kg	0.58	0.69	0.72	0.82	0.78	0.77	0.03	<0.001	0.003
ADFI, kg	1.49	1.66	1.61	1.79	1.68	1.63	0.06	0.086	0.029
G:F	0.390	0.418	0.443	0.460	0.463	0.473	0.013	<0.001	0.143
Experimental infection									
ADG, kg	0.55	0.58	0.72	0.78	0.73	0.75	0.03	<0.001	0.007
ADFI, kg	1.23	1.23	1.46	1.54	1.41	1.50	0.05	<0.001	0.037
G:F	0.445	0.468	0.495	0.503	0.515	0.500	0.018	0.011	0.147
Overall ⁶									
Control									
End BW, kg	116.2	130.2	123.4	130.7	131.5	125.2	2.49	0.016	0.006
ADG, kg	0.88	1.01	0.95	1.03	1.03	0.98	0.02	0.002	0.003
ADFI, kg	1.66	1.85	1.78	1.97	1.85	1.80	0.07	0.120	0.032
G:F	0.530	0.548	0.535	0.520	0.560	0.543	0.016	0.529	0.812
PRRSV									
End BW, kg	121.4	120.9	129.3	129.9	129.7	132.5	2.62	0.002	0.473
ADG, kg	0.90	0.90	0.97	0.97	0.97	1.01	0.02	0.001	0.712
ADFI, kg	1.44	1.41	1.66	1.72	1.61	1.69	0.05	<0.001	0.048
G:F	0.625	0.638	0.585	0.563	0.610	0.598	0.016	0.091	0.081

¹n = 4 pens per diet, 7 to 8 pigs per pen.

²Linear and quadratic orthogonal contrast.

³Pre-challenge adaptation period (-14 to 0 dpi), all pigs on common diet.

⁴Challenge period 1 (0 to 21 dpi), pigs fed experimental diets.

⁵Challenge period 2 (0 to 49 dpi), Control barn naturally infected with PRRSV after 21 dpi, pigs fed experimental diets.

⁶Overall challenge period (0 dpi to market); control pigs naturally infected after 21 dpi, PRRSV pigs experimentally infected at 0 dpi.

and feed efficiency were evaluated from 0 to 49 dpi to determine the effect of natural vs. experimental infection. In pigs naturally infected

with PRRSV, ADG increased linearly from 2.33 to 3.24 with increasing Lys:ME, resulting in both linear and quadratic effects ($P < 0.001$

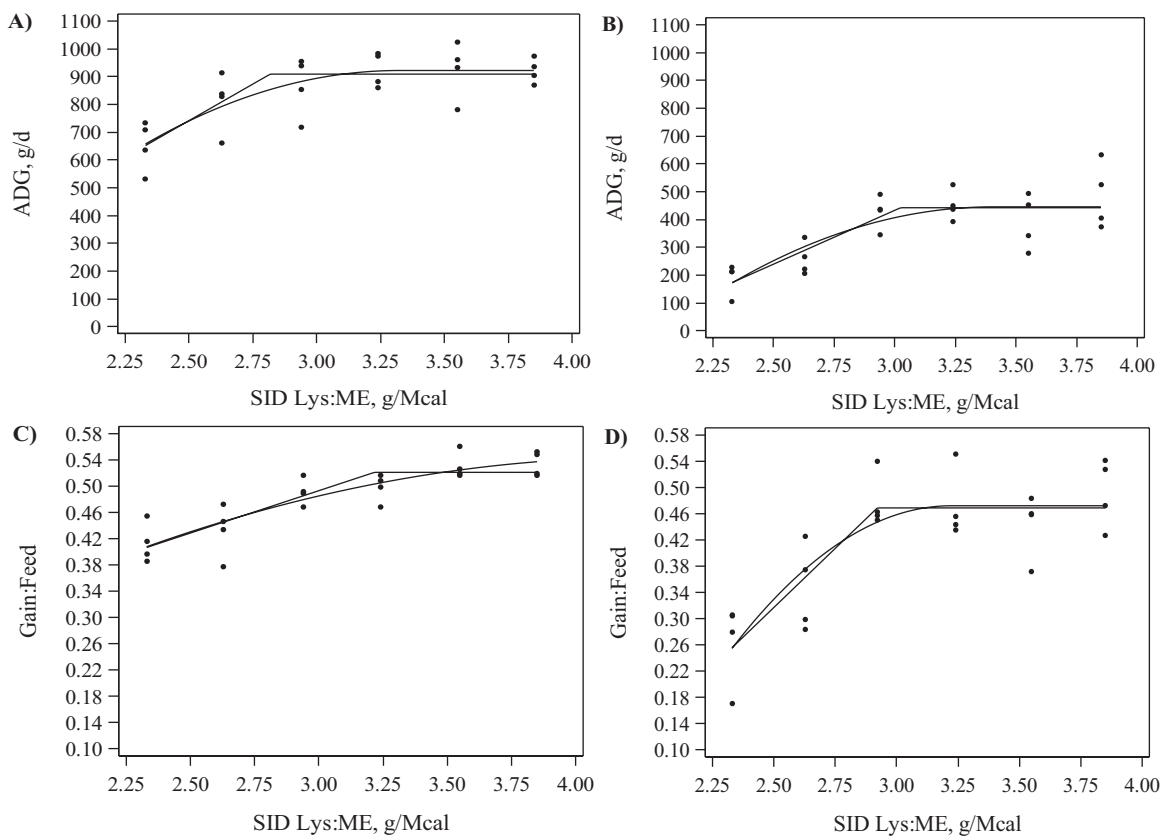


Figure 2. Data points represent treatment means from four pens per experimental diet per health status (Exp. 2). One-slope straight broken-line and quadratic broken-lines were fitted for maximum ADG (A and B) and G:F (C and D) expressed as a function of standardized ileal digestible (SID) Lys:ME (g SID Lys per Mcal ME) over a 21-d growth period in control (A,C) and PRRSV (B,D) infected pigs, respectively. (A) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 2.82 g/Mcal (Y plateau = 908.8 ADG; slope below requirement = -526.7 ; $r^2 = 0.59$). The quadratic broken-line model resulted in a SID Lys:ME requirement of 3.32 g/Mcal ($Y = 921.9 - 267.2(3.32 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.61$). (B) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 3.02 g/Mcal (Y plateau = 442.2 ADG; slope below requirement = -387.3 ; $r^2 = 0.65$). The quadratic broken-line model yielded a SID Lys:ME requirement of 3.41 g/Mcal ($Y = 445.4 - 235.8(3.41 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.63$). (C) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 3.22 g/Mcal (Y plateau = 0.521 G:F; slope below requirement = -0.129 ; $r^2 = 0.74$). The quadratic broken-line model yielded a SID Lys:ME requirement of 4.29 g/Mcal ($Y = 0.544 - 0.036(4.29 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.78$); however, this predicted value is outside the range of the experimental diets tested. (D) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 2.92 g/Mcal (Y plateau = 0.469 G:F; slope below requirement = -0.361 ; $r^2 = 0.72$). The quadratic broken-line model projected a SID Lys:ME requirement of 3.22 g/Mcal ($Y = 0.472 - 0.272(3.22 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.69$).

and $P = 0.003$, respectively). Also, in naturally infected pigs, ADFI increased quadratically ($P = 0.029$) with a peak at 3.24 Lys:ME and G:F increased linearly ($P < 0.001$) and with Lys:ME. From 0 dpi to market (approximately 100 d), ADG increased as Lys:ME increased, causing an increase in final BW (linear $P < 0.02$, quadratic $P < 0.01$, both parameters). Overall feed intake increased in a quadratic manner ($P = 0.032$). Breakpoint analysis determined 2.85 and 3.41 Lys:ME for optimal ADG using one-slope and quadratic broken-line models, respectively (Figure 3). Optimal G:F Lys:ME was achieved at 3.18 and 3.85 in one-slope and quadratic broken-line models, respectively (Figure 3).

Pigs experimentally infected with PRRSV demonstrated a similar response to increasing Lys:ME, with ADG and ADFI having a linear ($P < 0.001$, both parameters) and quadratic ($P = 0.007$ and $P = 0.037$, respectively) response, whereas G:F responded linearly to increasing Lys:ME ($P = 0.011$). Overall, final BW and ADG increased linearly with Lys:ME (linear $P \leq 0.002$, both parameters). Feed intake increased from 2.33 to 3.24 Lys:ME then decreased, leading to a linear ($P < 0.001$) and quadratic effect ($P = 0.048$). Optimal ADG was achieved at 3.12 and 3.47 Lys:ME using one-slope and quadratic broken-line breakpoint analysis, respectively. Optimal G:F was achieved at 3.08 and 3.52 Lys:ME using one-slope and quadratic broken-line models, respectively.

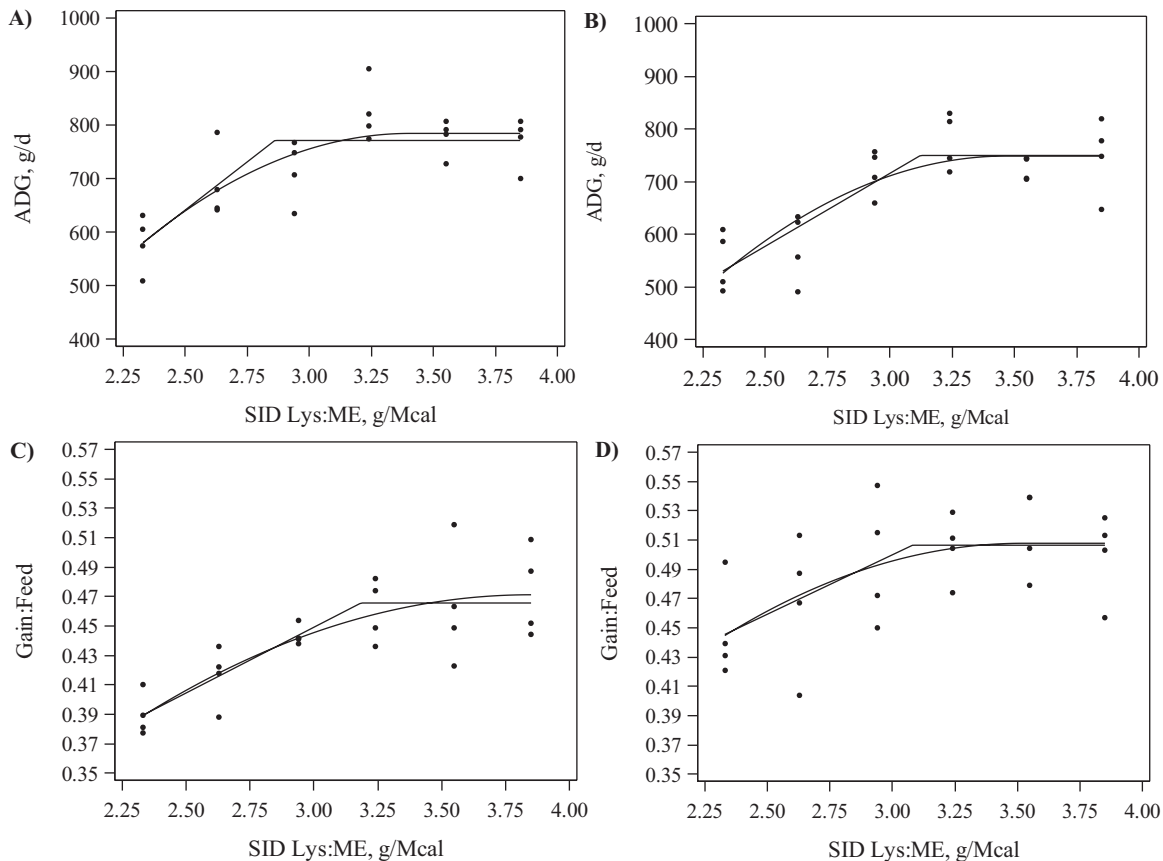


Figure 3. Data points represent treatments means from four pens per experimental diet per health status (Exp. 2). One-slope straight broken-line and quadratic broken-lines were fitted for maximum ADG (A and B) and G:F (C and D) expressed as a function of standardized ileal digestible (SID) Lys:ME (g SID Lys per Mcal ME) over a 49-d growth period in pigs that experienced a natural (A,C) or experimental (B,D) PRRSV infection, respectively. (A) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 2.86 g/Mcal (Y plateau = 771.3 ADG; slope below requirement = -360.0 ; $r^2 = 0.62$). The quadratic broken-line model resulted in a SID Lys:ME requirement of 3.41 g/Mcal ($Y = 784.4 - 176.4(3.41 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.67$). (B) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 3.12 g/Mcal (Y plateau = 749.6 ADG; slope below requirement = -276.8 ; $r^2 = 0.71$). The quadratic broken-line model yielded a SID Lys:ME requirement of 3.47 g/Mcal ($Y = 748.6 - 171.3(3.47 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.67$); however, this predicted requirement is outside the range of the diets tested. (C) The one-slope straight broken-line model yielded a SID Lys:ME requirement of 3.18 g/Mcal (Y plateau = 0.466 G:F; slope below requirement = -0.089 ; $r^2 = 0.64$). The quadratic broken-line model yielded a SID Lys:ME requirement of 3.85 g/Mcal ($Y = 0.471 - 0.036(3.85 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.65$). (D) The one-slope straight broken-line model resulted in a SID Lys:ME requirement of 3.08 g/Mcal (Y plateau = 0.506 G:F; slope below requirement = -0.081 ; $r^2 = 0.36$). The quadratic broken-line model projected a SID Lys:ME requirement of 3.52 g/Mcal ($Y = 0.508 - 0.044(3.52 - \text{g SID Lys/Mcal})^2$; $r^2 = 0.36$).

DISCUSSION

In healthy growing pigs, Lys is the first limiting AA for growth, and recommendations for Lys requirements have been widely established (NRC, 2012). Interestingly, when pigs are housed under unsanitary conditions, the Lys requirement for growth is reduced (Williams et al., 1997b, 1997c), which has been attributed to a reduced capacity for protein accretion (Williams et al., 1997a); however, efficiency of Lys utilization may not be different between healthy and immune-stimulated pigs. Therefore, reduced feed intake, and thus Lys intake, likely contributes to the reduction in lean tissue accretion and growth. In a similar unsanitary model, van der Meer et al. (2016) reported an

improvement in feed efficiency when Met, Thr, and Trp were increased 20% relative to Lys. In contrast, when immune system activation was modeled using repeated lipopolysaccharides (LPS), Met+Cys requirement was reduced (Rakhshandeh et al., 2014), but Met:Met+Cys requirement for protein deposition increased (Litvak et al., 2013). These data suggest that AA requirements in a model that mimics inflammation may be different from that of healthy pigs. Therefore, we conducted two experiments to determine how increasing Lys:ME impacted growth performance in healthy and PRRSV challenge pigs.

To the best of our knowledge, this is the first set of experiments to determine the Lys:ME requirements for optimal ADG and G:F in pigs challenged with PRRSV. Compared to healthy cohorts

in Exp. 1, PRRSV increased Lys:ME requirement for ADG by 21% to 36% depending on the statistical model used; however, the quadratic model predicted Lys:ME requirement to be 3.71 Lys:ME which is above the 3.41 Lys:ME tested in the study suggesting that the requirement could be higher than the test diets. Similarly, PRRSV increased the Lys:ME for optimal G:F by 11% to 30%. Similar to the predicted quadratic requirement for ADG, the G:F prediction was above the 3.41 Lys:ME diet and, therefore, the requirement may be higher than the tested diets. In Exp. 2, and in agreement with Exp. 1, optimal ADG was achieved at 3% to 7% higher Lys:ME in PRRSV-infected pigs compared to healthy controls. The PRRSV decreased Lys:ME requirement to achieve optimal G:F by 9% to 25%; however, optimal G:F in control pigs using a quadratic model predicted a requirement above the diets tested. Because control pigs in Exp. 2 became infected with PRRSV, the optimal Lys:ME was able to be determined for natural vs. experimental PRRSV infection. The optimal ADG and G:F was achieved at slightly higher Lys:ME levels in naturally infected pigs compared to experimentally infected cohorts. These data contrast with the classic articles by Williams et al. (1997a, 1997b, 1997c) that show Lys requirements to be less in immune-stimulated pigs compared to healthy pigs; however, Lys efficiency was not different between groups, suggesting growth differences are related to feed intake and Lys intake. A similar response occurs in broilers challenged with LPS, where Lys utilization by muscle does not change, but Lys utilization by the immune system increases 6-fold (Klasing and Calvert, 1999). As mentioned previously, soybean meal was used to increase dietary Lys, therefore, intake of other AA are likely increased. Acute-phase protein synthesis requires a large portion of aromatic AA (Reeds et al., 1994). Also, increased Met and Met + Cys can be beneficial to protein deposition in LPS-challenged pigs (Litvak et al., 2013; Rakhshandeh et al., 2014). Altogether, increased intake of these AA and others can reduce the need for lean tissue catabolism and preserve lean tissue and therefore growth.

In Exp. 1, although ADG was different between control and PRRSV-infected pigs, growth was optimized at total Lys intake of 22 and 21.5 g per day for control and PRRSV-infected pigs, respectively, and both were close to the recommended 20.5 g per day total Lys intake for 50 to 75 kg pigs by the NRC (2012). Although growth was different, it was optimized at similar Lys intake, which is somewhat similar to results from Williams et al. (1997b, 1997c)

where growth was similar at similar Lys intake; however, these two studies used an unsanitary environment challenge model not a live virus. In Exp. 2, 0 to 21 dpi ADG in control and PRRSV-infected pigs was maximized at 18.7 and 10.6 g per day total Lys intake, respectively. Control pigs had Lys intakes were close to the predicted 16.9 g Lys per day intake recommended by the NRC (2012) for 25 to 50 kg pigs. Although PRRSV-infected pigs were well below NRC (2012) recommendation total Lys intake, PRRSV-infected pigs had Lys intakes close to the 12.8 g per day Lys intake reported by Williams et al. (1997c) for optimal growth in 25 kg pigs raised under immune-stimulated, unsanitary environmental conditions. Infection appeared more severe in Exp. 2 as compared to Exp. 1, likely because pigs were younger, therefore, more severe infection could result in Lys efficiency differences.

When pigs are experimentally infected with a pathogen, the population is on the same disease plane as opposed to a natural infection that can lead to persistent, recurring infection (Yoon et al., 1999; Chand et al., 2012). Pigs that experienced natural and experimental PRRSV infection reached a similar peak viremia based on Ct values, and similar peak antibody. Naturally infected pigs appear to have experienced a shorter viremia duration; however, the same pigs were not bled for the duration of the growth period to more accurately determine PRRSV and antibody dynamics. Pigs became naturally infected around 45 kg, which likely allowed them to cope better with disease. As mentioned, pigs naturally and experimentally infected with PRRSV had a similar Lys:ME for optimal growth. Therefore, it is difficult to elucidate the effect of BW or diet on a potential protective role of against chronic PRRSV infection.

In the U.S. swine industry, soybean meal is primarily used to increase Lys and essential AA concentrations in the diet. Feeding increased soybean meal levels to PRRSV infected pigs may also have potential benefits for PRRSV viral clearance (Rochell et al., 2015); however, Lys:ME was not different between diets. In the study presented herein, altering Lys:ME by increasing soybean meal content of the diets did not alter viral titers, PRRSV genomic content or antibody response within the PRRSV-challenged pigs (Exp. 1). Although contrary to Rochell et al. (2015), this result is consistent with a previous study from our group (Schweer et al., 2018). Increased dietary soybean meal, regardless of Lys:ME can also increase performance in finishing pigs naturally infected with PRRSV and porcine circovirus disease (Boyd et al., 2010). In 8 kg pigs infected with

PRRSV, ADG was also improved in a soybean meal diet vs. a soybean meal plus crystalline AA diet with the same Lys:ME (Rochell et al., 2015). Although a linear increase in ADG and G:F has been reported in 55 kg pigs that were PRRSV positive at weaning as SID lysine increases (Shelton et al., 2011). In agreement with these reports, pigs infected with PRRSV in this study showed linear improvements in ADG and G:F as Lys:ME increased.

In agreement with Li et al. (2012), fat thickness was impacted by Lys:ME in control pigs. In this study, there was a clear quadratic effect while Li et al. demonstrated both linear and quadratic effects; however, in this study experimental diets were not fed up until carcass data were collected as was the case in the study performed by Li et al. (2012). Interestingly, when comparing fat thickness in control and PRRSV-infected pigs, there is an opposite effect of Lys:ME, where fat thickness decreased from 1.87 to 2.80 Lys:ME in control pigs and then increased up to 3.41 Lys:ME. In PRRSV-infected pigs, fat depth increased linearly with increasing Lys:ME. Our group has shown that fat accretion is decreased in PRRSV-infected pigs (Schweer et al., 2017), therefore, increasing the AA profile of the diet during a PRRSV challenge period may aid in maintaining energy levels and therefore body fat. In Exp. 1, although growth rates were different during the challenge period control and PRRSV-infected pigs reached finishing weight at the same time, suggesting the possibility of compensatory growth. Compensatory growth is a phenomenon where pigs accelerate growth after a period of feed or nutrient restriction, although this is not consistently observed (Mersmann et al., 1987; Taylor et al., 2013).

In summary, increased Lys:ME during a 35 d or 21 d PRRSV challenge in 50 and 25 kg pigs, respectively, increases ADG and G:F. There was no difference in immune response, as determined by PRRS viremia or antibody response, and no difference in carcass characteristics. When break-point analysis was performed in Exp. 1, optimal Lys:ME for ADG and G:F was increased up to 36% and 30%, respectively, in PRRSV-infected pigs compared to healthy controls. In Exp. 2, optimal Lys:ME for ADG increased up to 7%; however, optimal Lys:ME for G:F was decreased up to 25% in PRRSV-infected pigs. In Exp. 1, the predicted requirement for ADG and G:F in PRRSV-infected pigs using a quadratic model were above the highest Lys:ME diet. This was similar for G:F in control pigs in Exp. 2, therefore, further studies should be conducted to more accurately determine the

Lys:ME requirement. In Exp. 2, it was also determined that Lys:ME for optimal ADG and G:F between pigs naturally and experimentally infected with PRRSV was not different. Altogether, increasing Lys:ME above the NRC requirement increased performance and feed efficiency in PRRSV-infected pigs, and the response was similar between natural and experimental PRRSV infection.

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