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Standardized ileal digestible lysine requirements of male pigs immunized against gonadotrophin releasing factor[1]

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ABSTRACT: An experiment was conducted to determine the standardized ileal digestible (SID) Lys requirement of entire male and male pigs immunized against gonadotrophin releasing factor (GnRF; immunocastrates). A total of 420 entire male and immunocastrated (IC) male pigs weighing 60.1 kg BW (SEM 0.49) were used in a 2 × 5 factorial experiment with the main effects being gender (entire males or IC males) and 5 concentrations of SID Lys:DE ratio (0.32, 0.43, 0.54, 0.64, or 0.75 g SID Lys/MJ DE). The diets were fed for 6 wk until slaughter at 107.5 kg BW (SEM 5.72). Over the entire period, IC males had a greater ADG ($P < 0.001$), greater ADFI ($P < 0.001$), and lower G:F ($P < 0.001$) compared with entire males. Immunocastrated males had increased plasma urea nitrogen (PUN) concentrations compared with entire males from d 10 to 42 ($P < 0.001$ for all days). Plasma urea nitrogen concentration also increased as Lys concentrations increased from d 3 to 42 ($P < 0.001$ for all days). Using the linear-plateau model, the optimal ADG for entire males was achieved at SID Lys concentrations of 0.68, 0.62, 0.54, and 0.58 g/MJ DE whereas optimal G:F was achieved at SID Lys concentrations of 0.72, 0.60, 0.54, and 0.51 g/MJ DE for the time periods d 0 to 14, d 15 to 28, d 29 to 42, and d 0 to 42, respectively. For IC males, optimal ADG was achieved at SID Lys concentrations of 0.64, 0.43, 0.38, and 0.40 g/MJ DE whereas optimal G:F was achieved at SID Lys concentrations of 0.64, 0.43, 0.36, and 0.42 g/MJ DE for the same respective time periods. Using the quadratic polynomial model, maximum ADG for entire males was achieved at SID Lys concentrations of 0.62 and 0.58 g/MJ DE whereas maximum G:F was achieved at SID Lys concentrations of 0.59 and 0.68 g/MJ DE for d 29 to 42 and d 0 to 42, respectively. For IC pigs, maximum ADG was achieved at SID Lys concentrations of 0.69, 0.54, and 0.64 g/MJ DE whereas maximum G:F was achieved at SID Lys concentrations of 0.81, 0.54, and 0.64 g/MJ DE for d 0 to 14, d 29 to 42, and d 0 to 42, respectively. A solution could not be found using the quadratic polynomial model for entire males for d 0 to 14 for both ADG and G:F and for both entire males and IC males for d 15 to 28 within the range of Lys values tested. When both the growth performance and PUN values are considered, the results suggest that IC males show a response to dietary SID Lys similar to that of entire males for 2 wk after the second immunization against GnRF. After this, IC males have a lower requirement for SID Lys than entire males.

Key words: immunocastrates, pig, response to lysine

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INTRODUCTION

Immunization of entire males against gonadotrophin releasing factor (**GnRF**) is an alternative to physical castration (Dunshea, 2009). It involves immunization with an incomplete analog of GnRF conjugated to a carrier protein in a low reactogenic–adjuvant system (Dunshea et al., 2001). The pig

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grows as an entire male with associated positive effects on growth and carcass leanness until the second immunization against GnRF, after which it becomes more similar to a physical castrate (Dunshea et al., 2001). It is crucial to provide the optimum standardized ileal digestible (**SID**) Lys concentration to pigs to ensure lean growth is maximized and feed costs are minimized (Main et al., 2008). There do not appear to be any dose–response studies in the literature that estimate the dietary Lys concentrations required by immunocastrated (**IC**) males to optimize growth performance (Millet et al., 2011; Dunshea et al., 2013). The nutrient requirements of IC males will be different from entire males and physical castrates as immunization against GnRF alters performance (Millet et al., 2011; Dunshea et al., 2013). In addition, Lys concentrations of IC males may change after the second immunization against GnRF as they become more similar to physical castrates, and so it is important that SID Lys concentrations for IC males are determined to benefit from the 2 distinct growth phases of IC males (Dunshea, 2009). The hypothesis was that IC male pigs will have a lower optimal SID Lys:megajoule (**MJ**) DE ratio than entire males beyond 2 wk after the second immunization against GnRF. The objectives of this experiment were to determine the SID Lys requirement of IC males compared with entire males and to examine how quickly Lys requirements change after the second immunization against GnRF.

MATERIALS AND METHODS

The experimental protocol used was approved by the Department of Agriculture and Food Western Australia's Animal Research Committee and by the Animal Ethics Committee (activity number 2-10-7). The animals were handled according to the Australian code of practice for the care and use of animals for scientific purposes (National Health and Medical Research Council, 2004).

A total of 420 (progeny of Large White × Landrace dams sired by Landrace × Duroc boars; Pig Improvement Company, Grong Grong, NSW, Australia) entire male and IC male pigs were used in this experiment. The experiment was a 2 × 5 factorial with the main treatments being gender (entire males or IC males) and 5 concentrations of SID Lys-to-DE ratio (0.32, 0.43, 0.54, 0.64, or 0.75 g SID Lys/MJ DE). There were 6 replicate pens per treatment with 7 pigs allocated per pen. The pigs weighed 60.1 kg BW (SEM 0.49) at the commencement of the experiment and were slaughtered 6 wk later at 107.5 kg BW (SEM 5.72).

Allocation and Housing

Pigs were sourced at approximately 50 kg live weight from a high health status commercial herd whose bloodlines were sourced from the Pig Improvement Company. Due to pig availability, the pigs were sourced in 6 equal batches. Upon arrival at the research station, the pigs were individually identified with ear tags, weighed, and stratified based on BW before being allocated to treatment. The pigs were housed at 0.93 m^2/pig in groups of 7 in a naturally ventilated grower–finisher facility. All pigs had ad libitum access to feed via a single-spaced feeder and water for the entire period of the experiment. Pigs received their priming dose of anti-gonadotrophin releasing factor immunological product (Improvac; Zoetis Australia, Rhodes, Australia) at approximately 10 wk of age on the farm. The second Improvac immunization was given to the designated pigs 6 wk before slaughter (60.1 kg BW [SEM 0.49). The entire males did not receive a placebo injection.

Diets

The pigs were fed a commercial diet from arrival until the commencement of the experimental diet. Two base experimental diets were formulated with DE contents of 13.5 and 13.6 MJ/kg and available Lys contents of 0.32 and 0.75 g SID Lys/MJ DE, respectively. With the exception of Lys, the diets were formulated to meet or exceed the requirements of the pigs (NRC, 1998; Shelton et al., 2011). The 2 diets were then blended in the appropriate proportions using a Feedlogic system (FEEDPro automated feed delivery system; Feedlogic Corp., Willmar, MN) to attain the desired available Lys contents. The composition of the diets and the ratios used to attain the blended diets are presented in Table 1. The feed samples were also analyzed for quantitative AA composition (Australian Proteome Analysis Facility, Sydney, NSW, Australia) and the results are presented in Table 2.

Slaughter Procedure

Six weeks after the diets were introduced, the pigs were individually tattooed, removed from feed overnight, and transported to a commercial abattoir (approximately 90 min transport time). The pigs were stunned using a carbon dioxide, dip-lift stunner set at 85% CO_2 for 1.8 min (Butina A/S, Holbaek, Denmark). Exsanguination, scalding, dehairing, and evisceration were performed using standard commercial procedures (Moore et al., 2009).

Table 1. The calculated composition of the basal diets for the 2 extreme Lys concentrations

Diet	Diet 1 (low)[1]	Diet 2 (high)
Ingredients, g/kg, as-fed basis		
Barley	517	100
Triticale	50.0	200
Wheat	255	235
Groats	20.0	100
Wheat mill run	20.0	31.7
Canola meal, 36%	10.0	100
Soybean meal, 48%	50.0	168
Meat meal	20.0	36.2
Tallow	32.9	10.0
L-Lysine	0	2.01
Methionine Alimet[2]	0	0.48
Threonine	0	0.80
Minerals and vitamins[3]	2.50	2.50
Limestone	9.21	6.22
Dicalium phosphorus	10.5	5.00
Salt	2.50	2.50
Choline chloride	0.20	0
Nutrient composition[4]		
SID[5] AA		
Lys, g/kg	4.30	10.2
Ile:Lys	9.00	0.70
Leu:Lys	1.67	1.24
Met:Lys	0.36	0.30
Met + Cys:Lys	0.85	0.60
Thr:Lys	0.79	0.68
Trp:Lys	0.26	0.20
Val:Lys	1.13	0.81
DE, MJ/kg	13.5	13.6
Total Lys, g/kg	5.40	12.0
CP, g/kg	123	210
Ca, g/kg	9.00	9.00
Total P, g/kg	6.20	7.10
Available P, g/kg	4.00	4.00
Na, g/kg	1.30	1.40
NDF, g/kg	156	143
ADF, g/kg	49.6	71.6
g SID Lys/MJ DE	0.32	0.75

[1]Ratios of Diet 1:Diet 2 were 100:0, 75:25, 50:50, 25:75, or 0:100 for 0.32, 0.43, 0.54, 0.64, and 0.75 g standardized ileal digestible Lys/MJ DE, respectively.

[2]Liquid methionine (88%; Novus International Inc., St. Charles, MO).

[3]Provided per kilogram of final diet: 7,000 IU vitamin A, 1,400 IU vitamin D_3, 20 g vitamin E, 1 g vitamin K, 1 g vitamin B_1, 3 g vitamin B_2, 1.5 g vitamin B_6, 15 mg vitamin B_{12}, 12 g niacin, 10 mg pantothentic acid, 0.19 g folic acid, 30 mg biotin, 10.6 g calcium pantothenatic, 60 g iron, 100 g zinc, 40 g manganese, 10 g copper, 0.2 g cobalt, 0.5 g iodine, 0.3 g selenium, and 20 g antioxidant.

[4]Calculated composition.

[5]SID = standardized ileal digestible.

Table 2. Quantitative AA analysis of the basal diets used

AA, g/kg, as-fed basis	Low	High
Histidine	2.90	4.90
Isoleucine	4.90	8.20
Leucine	9.10	14.5
Lysine[1]	5.60	11.6
Methionine	1.60	2.30
Phenylalanine	5.90	9.10
Threonine	4.60	8.10
Valine	6.40	10.2

[1]Based on the analyzed concentrations for the 2 extreme diets, the estimated Lys concentrations for all diets are 5.6, 7.1, 8.6, 10.1 and 11.6 g/kg, respectively.

Growth Performance and Carcass Composition Assessment

Pigs were weighed weekly, and feed intake was determined during each weigh period for calculation of ADG, ADFI, and G:F. Hot carcass weight (AUS-MEAT Trim 13; AUS-MEAT Ltd., South Brisbane, QLD, Australia) and P2 backfat depth, 65 mm from the dorsal midline at the point of the last rib (Pork-Scan Pty. Ltd., Canberra, Australia), were measured approximately 35 min after exsanguination, before chiller entry (2°C and 4 m/s airspeed).

Blood Analysis

Blood samples (20 mL in lithium heparin tubes) were collected from all pigs in the pen of every second batch (n = 21 pigs) on d 0 (when experimental diets commenced and the second immunization against GnRF was given), 3, 7, 10, 14, 21, 28, 35, and 42. The blood samples were centrifuged at 2,000 × g for 10 min at 5°C to recover plasma and were stored at −20°C until analyzed. Plasma urea nitrogen (**PUN**) was quantified using a commercial kit (Olympus kit catalog number OSR6134; ±0.347 at 7.75 mmol/L; Olympus UK Ltd., Hertfordshire, UK) and the assay was performed on an automated analyzer according to the manufacturer's instructions (Olympus AU400; Olympus UK Ltd.). Plasma urea nitrogen (mmol/L) was converted to PUN (mg/dL) by dividing by 0.357. Plasma testosterone was measured using an extraction titrated RIA on d 0, 14, and 42 (2 pigs per pen in every second batch; n = 30 per gender). Coefficients of variation within ssays were 6.4 (2.58 ng/mL), 6.3 (0.98 ng/mL), and 7.3% (0.35 ng/mL).

Statistical Analysis

Two-way ANOVA was performed with the GenStat 14 program (VSN International Ltd., Hemel Hempstead, UK) to analyze the main effects of gender and Lys. The response to Lys was tested for linear and

quadratic effects using polynomial orthogonal contrasts. Arrival batch was used as a block in the analysis and pen was used as the experimental unit. Data for testosterone and PUN were analyzed using a square root transformation to ensure model assumptions were not violated. The testosterone data were pooled across Lys concentration and analyzed as repeated measures (GenStat 14) with gender as the main effect. A level of probability of less than 0.05 was used to determine statistical difference between the means. A level of probability of less than 0.1 was determined to be a trend. Fisher's protected LSD were used to determine differences among treatments.

Quadratic curves were fitted to the treatment means to predict the optimum dietary Lys concentration for maximum daily gain and G:F for the periods 0 to 14 d (d 0–14), 15 to 28 d (d 15–28), 29 to 42 d (d 29–42), and 0 to 42 d (d 0–42) after the second immunization against GnRF. The quadratic curves were fitted using $y = ax^2 + bx + c$, in which y = either daily gain or G:F, x = g SID Lys/megajoule DE, and a, b, and c are representative components of the equation (O'Connell et al., 2006). The data were also analyzed using the linear plateau model fitted to the treatment means. The model was y = maximum + rate constant × (requirement − x) if $x \leq$ requirement or y = maximum if $x >$ requirement, in which x = g SID Lys/MJ DE (Nutrient Response Models Version 1.1 [University of Georgia, Athens, GA]; Excel [Microsoft Corporation, Redmond, WA]; Vedenov and Pesti, 2008).

RESULTS

The analyzed AA values are given in Table 2. The variation between calculated and analyzed Lys values for the low-Lys diet was 4% whereas for the high-Lys diet, it was 3%. The difference between formulated and actual values is within the permitted analytical variation of 20% for Lys analysis accepted by the Association of American Feed Control Officials (2005, as cited in Shelton et al., 2011).

Immunocastrates had a greater ADG and a greater ADFI from d 15 to 28, from d 29 to 42, and from d 0 to 42 (all $P < 0.001$) compared with entire males (Fig. 1 and 2). Gain-to-feed ratio was better for entire males from d 29 to 42 ($P = 0.007$) and from d 0 to 42 ($P < 0.001$) compared with IC males (Fig. 3). Immunocastrates were heavier on d 42 ($P < 0.001$) and had a heavier HCW ($P < 0.001$), a lower dressing percentage ($P < 0.001$), and increased backfat ($P < 0.001$) compared with the entire males (Table 3).

Increasing the concentration of Lys increased both ADG and G:F for all time periods ($P < 0.001$ and $P < 0.001$, respectively; Fig. 1 and 3). Average daily feed intake also increased as the concentration of Lys increased for d 0 to 14, d 15 to 28, d 29 to 42, and d 0 to 42 ($P = 0.039$, $P = 0.029$, $P = 0.003$, and $P = 0.005$, respectively; Fig. 2). Pigs that received the lower concentrations of Lys had a lower HCW ($P < 0.001$) and lower dressing percentage ($P = 0.007$) and increased backfat ($P < 0.001$) compared with pigs that received the higher concentrations of Lys (Table 3).

The interaction showed that the IC males had a greater ADG at lower Lys concentrations compared with entire males for d 29 to 42 and d 0 to 42 ($P < 0.001$ and $P < 0.001$, respectively). There was also a trend for this to occur for ADG for d 0 to 14 and d 15 to 28 ($P = 0.076$ and $P = 0.069$, respectively). At higher concentrations of Lys, IC males had a lower G:F compared with entire males from d 29 to 42 and from d 0 to 42 ($P < 0.001$ and $P < 0.001$, respectively). There was also a trend for this to occur for G:F for d 0 to 14 and d 15 to 28 ($P = 0.055$ and $P = 0.060$, respectively).

There was a positive linear effect of Lys for both ADG and G:F ($P < 0.05$) for entire and IC males for all time periods with the exception of d 29 to 42 for IC males ($P > 0.05$; Fig. 1 and 3). Both ADG and G:F quadratically improved with increasing Lys ($P < 0.05$) for both entire males and IC males for all time periods with the exception of d 0 to 14 and d 15 to 28 for entire males and d 15 to 28 for IC males. For ADFI, there was a positive linear effect of increasing Lys concentration ($P < 0.05$) for entire males for all time periods ($P < 0.05$) but no effect for IC males at any time (Fig. 2). There were no quadratic effects of Lys ($P > 0.05$) for ADFI for either entire males or IC males at any time. Hot carcass weight increased both linearly ($P < 0.001$ and $P = 0.024$) and quadratically ($P < 0.001$ and $P < 0.001$) with increasing Lys concentration for entire males and IC males, respectively. Increasing Lys concentration linearly increased dressing percentage ($P < 0.001$) for IC males and quadratically increased dressing percentage for both entire males ($P < 0.001$) and IC males ($P < 0.001$). Backfat linearly decreased ($P < 0.001$ and $P < 0.001$, respectively) as the Lys concentration increased for both entire males and IC males (Table 3).

Immunocastrated males had an increased PUN concentration compared with entire males from d 10 to 42 ($P < 0.001$ for d 10, 14, 21, 28, and 35 and $P = 0.01$ for d 42; Fig. 4). Plasma urea nitrogen concentration also increased as the concentration of Lys increased from d 3 to 42 ($P < 0.001$ for all days except d 7 [$P = 0.05$]).

There was no difference between IC males and entire males for plasma testosterone concentrations on d 0 ($P > 0.05$; Fig. 5). Testosterone concentrations were lower for IC males on both d 14 and 42 compared with entire males ($P < 0.001$ and $P < 0.001$, respectively).

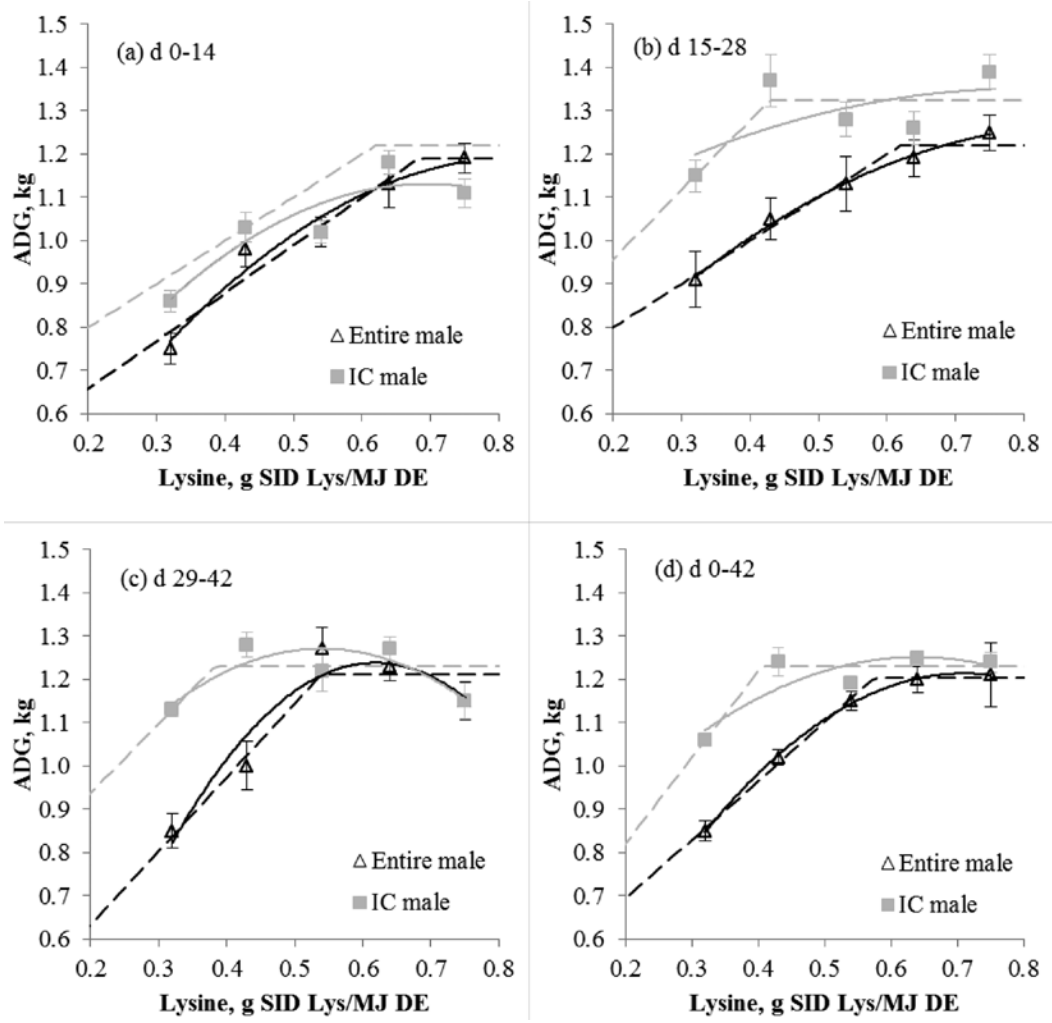

Figure 1. Average daily gain for entire males (triangle) and immunocastrated (IC) males (shaded square) fed varying concentrations of Lys (g standardized ileal digestible [SID] Lys/MJ DE) from 60.1 to 107.5 kg ($n = 6$) for (a) d 0 to 14, (b) d 15 to 28, (c) d 29 to 42, and (d) d 0 to 42. Treatment means are fitted with a linear plateau (- - -) and quadratic polynomial (—) model for each gender. (a) For d 0 to 14, the SEM for entire males and IC males was 0.057 and 0.042, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P = 0.087$, respectively, whereas for IC males, it was $P < 0.001$ and $P = 0.007$, respectively. (b) For d 15 to 28, the SEM for entire males and IC males was 0.075 and 0.062, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P = 0.353$, respectively, whereas for IC males, it was $P = 0.023$ and $P < 0.001$, respectively. (c) For d 29 to 42, the SEM for entire males and IC males was 0.064 and 0.048, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P < 0.001$, respectively, whereas for IC males, it was $P = 0.735$ and $P = 0.003$, respectively. (d) For d 0 to 42, the SEM for entire males and IC males was 0.036 and 0.031, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P < 0.001$, respectively, whereas for IC males, it was $P < 0.001$ and $P = 0.002$, respectively.

Plasma testosterone concentrations were lower for IC males on d 14 and 42 than d 0 ($P < 0.001$) with no difference between d 14 and 42 whereas entire males had lower plasma testosterone concentrations on d 0 and 42 compared with d 14 ($P < 0.001$). There was no effect ($P > 0.05$) of dietary Lys on plasma testosterone concentrations (data not shown).

The data were analyzed with a quadratic polynomial model and a linear-plateau model to determine the SID Lys requirement for optimal ADG and G:F for the time periods d 0 to 14, d 15 to 28, d 29 to 42, and d 0 to 42. The average starting BW for each of the periods d 0 to 14, d 15 to 28, d 28 to 42, and d 0 to 42 was 60.1 (SEM 0.49), 74.6 (SEM 0.63), 91.3 (SEM 0.93), and 60.1 (SEM 0.49) kg, respectively. Using the linear-plateau model, the breakpoint estimate to optimize ADG for entire males was achieved at SID Lys concentrations of 0.68, 0.62, 0.54, and 0.58 g/MJ DE whereas optimal G:F was achieved at SID Lys concentrations of 0.72, 0.60, 0.54, and 0.51 g/MJ DE for the respective time periods. For IC males, however, optimal ADG was achieved at SID Lys concentrations of 0.64, 0.43, 0.38, and 0.40 g/MJ DE whereas optimal G:F was achieved at SID Lys concentrations of 0.64, 0.43, 0.36, and 0.42 g/MJ DE for the respective time periods.

Using the quadratic polynomial model, maximum ADG for entire males was achieved at SID Lys concentrations of 0.62 and 0.58 g/MJ DE whereas maximum G:F was achieved at SID Lys concentrations of 0.59 and 0.68 g/MJ DE for d 29 to 42 and d 0 to 42, respectively.

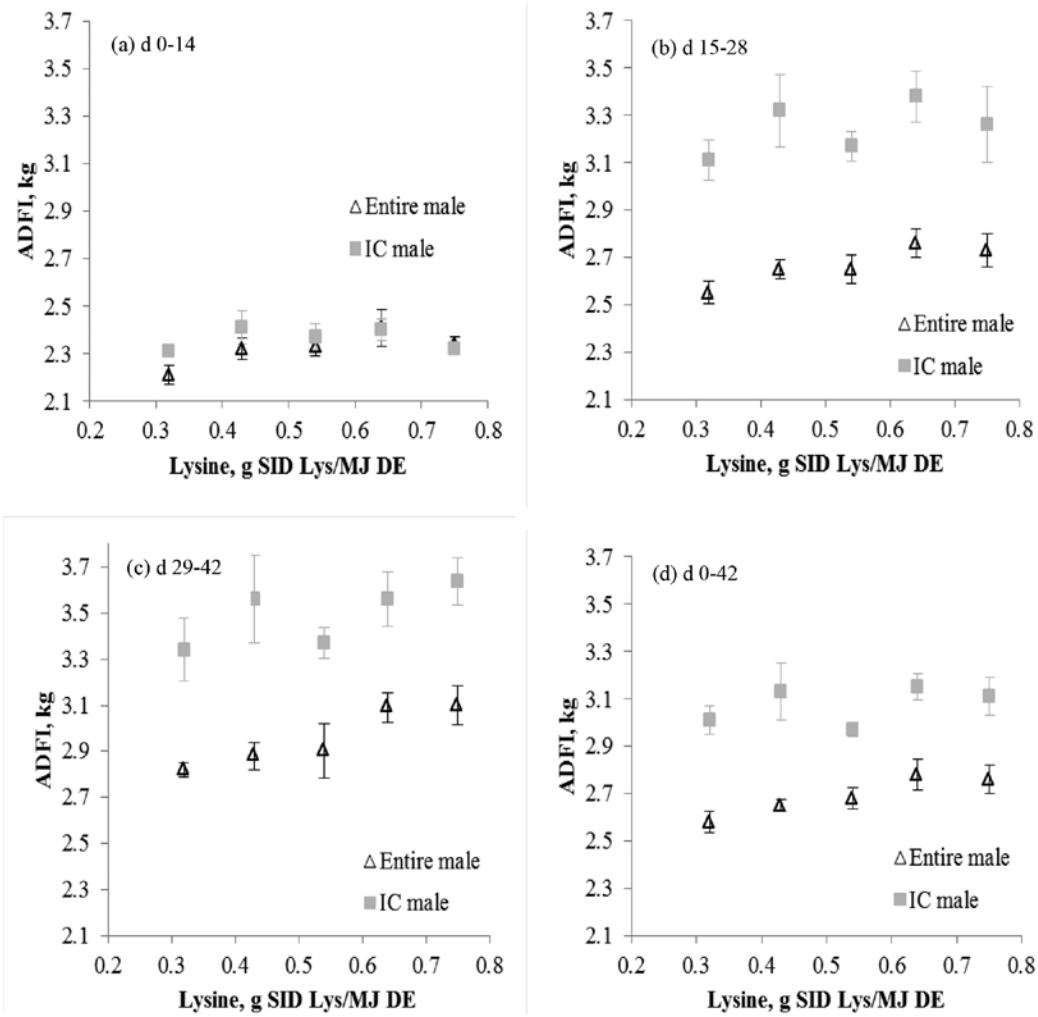

Figure 2. Average daily feed intake for entire males (triangle) and immunocastrated (IC) males (shaded square) fed varying concentrations of Lys (g standardized ileal digestible [SID] Lys/MJ DE) from 60.1 to 107.5 kg ($n = 6$) for (a) d 0 to 14, (b) d 15 to 28, (c) d 29 to 42, and (d) d 0 to 42. (a) For d 0 to 14, the SEM for entire males and IC males was 0.070 and 0.066, respectively. The linear and quadratic within-gender Lys effect for entire males was $P = 0.034$ and $P = 0.125$, respectively, whereas for IC males, it was $P = 0.916$ and $P = 0.112$, respectively. (b) For d 15 to 28, the SEM for entire males and IC males was 0.079 and 0.170, respectively. The linear and quadratic within-gender Lys effect for entire males was $P = 0.013$ and $P = 0.465$, respectively, whereas for IC males, it was $P = 0.316$ and $P = 0.497$, respectively. (c) For d 29 to 42, the SEM for entire males and IC males was 0.108 and 0.184, respectively. The linear and quadratic within-gender Lys effect for entire males was $P = 0.004$ and $P = 0.828$, respectively, whereas for IC males, it was $P = 0.159$ and $P = 0.834$, respectively. (d) For d 0 to 42, the SEM for entire males and IC males was 0.070 and 0.107, respectively. The linear and quadratic within-gender Lys effect for entire males was $P = 0.005$ and $P = 0.515$, respectively, whereas for IC males, it was $P = 0.388$ and $P = 0.949$, respectively.

For IC pigs, however, maximum ADG was achieved at SID Lys concentrations of 0.69, 0.54, and 0.64 g/MJ DE whereas maximum G:F was achieved at SID Lys concentrations of 0.81, 0.54, and 0.64 g/MJ DE for d 0 to 14, d 29 to 42, and d 0 to 42, respectively. A solution could not be found using the quadratic polynomial model for entire males for d 0 to 14 for both ADG and G:F and for both entire males and IC males for d 15 to 28 within the range of Lys values tested (Table 4).

DISCUSSION

Results from the current experiment show that the hypothesis that IC male pigs will have a lower optimal SID Lys:MJ DE ratio than entire males beyond 2 wk after the secondary immunization can be supported. For the first 2-wk period after the second immunization against GnRF, the predicted SID Lys concentration to optimize ADG and G:F was only slightly lower for the IC males compared with the entire males. Although there was some difference in the predicted Lys requirement to maximize ADG and G:F depending on the model used, the similarity in PUN concentrations between entire males and IC males provides further support that they show a similar response to Lys up to approximately d 10 after the second immunization against GnRF. Beyond this, PUN concentrations are greater for IC males compared with entire males. From d 15 to 28 after the second immunization against GnRF, there was a considerable difference in the SID Lys concentrations estimated to optimize ADG and G:F for entire males and IC males. Using only the

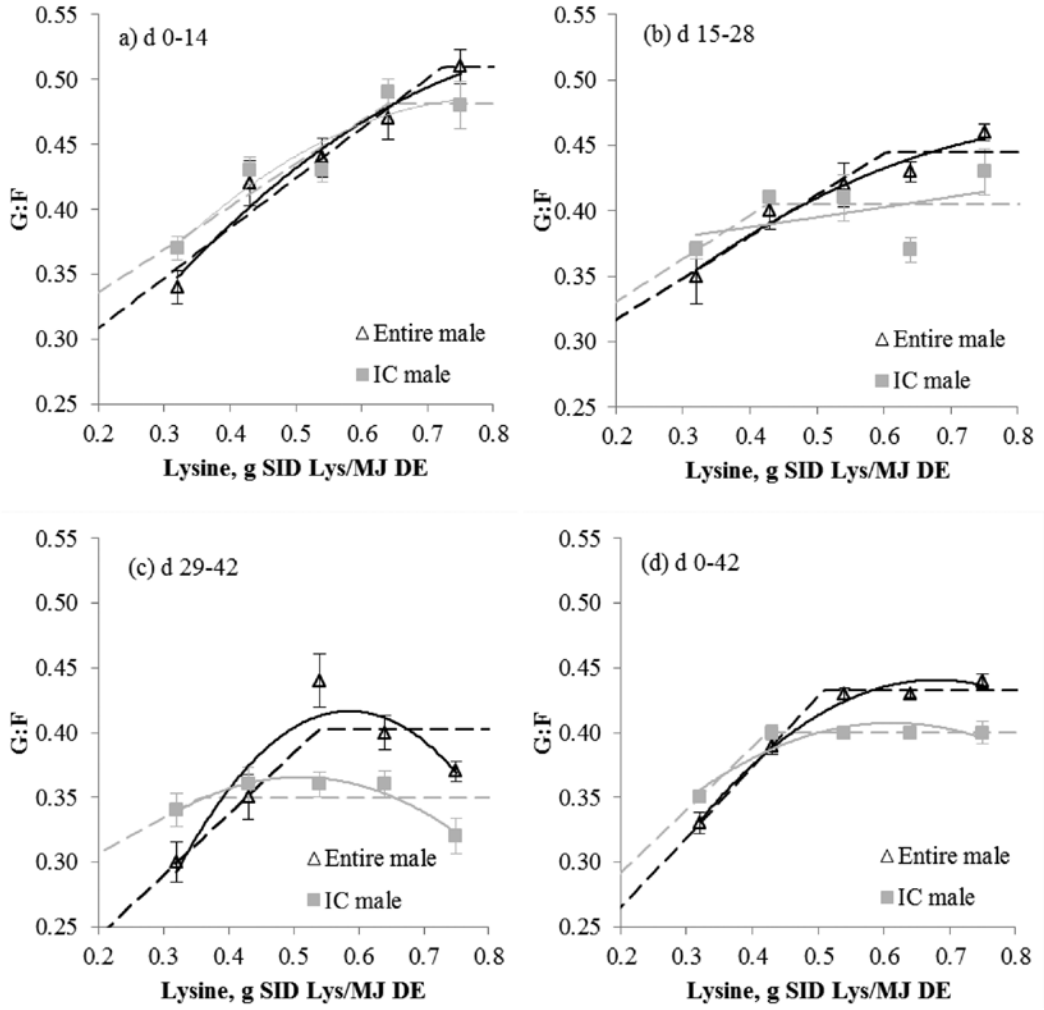

Figure 3. Gain-to-feed ratio for entire males (triangle) and immunocastrated (IC) males (shaded square) fed varying concentrations of Lys (g standardized ileal digestible [SID] Lys/MJ DE) from 60.1 to 107.5 kg ($n = 6$) for (a) d 0 to 14, (b) d 15 to 28, (c) d 29 to 42, and (d) d 0 to 42. Treatment means are fitted with a linear plateau (- - -) and quadratic polynomial (—) model for each gender. (a) For d 0 to 14, the SEM for entire males and IC males was 0.021 and 0.016, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P = 0.207$, respectively, whereas for IC males, it was $P < 0.001$ and $P = 0.073$, respectively. (b) For d 15 to 28, the SEM for entire males and IC males was 0.020 and 0.017, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P = 0.349$, respectively, whereas for IC males, it was $P = 0.027$ and $P = 0.858$, respectively. (c) For d 29 to 42, the SEM for entire males and IC males was 0.022 and 0.017, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P < 0.001$, respectively, whereas for IC males, it was $P = 0.209$ and $P = 0.009$, respectively. (d) For d 0 to 42, the SEM for entire males and IC males was 0.008 and 0.008, respectively. The linear and quadratic within-gender Lys effect for entire males was $P < 0.001$ and $P < 0.001$, respectively, whereas for IC males it was $P < 0.001$ and $P < 0.001$, respectively.

linear-plateau model, the optimal ADG and G:F were achieved at SID Lys concentrations of 0.69 and 0.60 g/MJ DE, respectively, for entire males whereas for IC males, these were 0.43 and 0.43 g/MJ DE, respectively. The predicted SID Lys requirements for both the entire males and IC males further declined for the period d 29 to 42; however, again, the SID Lys requirements were lower for the IC males compared with the entire males. The decrease in the Lys requirement beyond 2 wk after the second immunization against GnRF is likely associated with the increased feed intake and decline in protein deposition after the secondary immunization (Dunshea et al., 2013).

Although the Lys requirements are presented in 2-wk periods following the second immunization against GnRF, it is acknowledged that some of the pigs are initially fed Lys-deficient diets and this may have impacted the subsequent requirement in the next period. However, this carry-over effect is expected to be relatively minimal due to the small periods over which the prediction of the Lys requirements was made.

There do not appear to be any dose–response studies in the literature that estimate AA requirements of IC males (Millet et al., 2011; Dunshea et al., 2013). Boler et al. (2011a,b) investigated the effects of increasing Lys on carcass composition, cutting yields, and processed products from IC males with the main goal to maximize carcass quality. In these experiments, the second immunization against GnRF was given at 91 kg BW and the experimental diets (0.7, 0.8, 0.9, and

Table 3. Final BW and carcass characteristics for entire male and immunocastrated (IC) male pigs fed varying concentrations of Lys (g standardized ileal digestible [SID] Lys/MJ DE) from 60.1 to 107.5 kg (n = 6)

	Lys concentration, g SID Lys/MJ DE						*P*-value			Within-gender Lys concentration effect *P*-value[2]	
Item	0.32	0.43	0.54	0.64	0.75	SEM[1]	Lys	G[3]	Lys × G[4]	Lin.	Quad.
BW Day 42, kg											
Entire male	95.4	102	108	110	110	1.34	<0.001	<0.001	<0.001	<0.001	<0.001
IC male	104	111	110	112	112					<0.001	0.103
HCW, kg											
Entire male	63.2	69.2	73.2	74.7	73.9	0.959	<0.001	<0.001	<0.001	<0.001	<0.001
IC male	68.4	73.8	74.3	75.0	74.3					0.024	<0.001
Dressing percentage											
Entire male	66.2	67.4	68.0	67.6	66.8	0.392	0.007	<0.001	0.483	0.177	<0.001
IC male	65.8	66.4	67.8	66.9	66.5					<0.001	<0.001
Backfat, mm											
Entire male	13.0	12.9	11.1	10.4	9.86	0.757	<0.001	<0.001	0.298	<0.001	0.714
IC male	15.1	13.2	12.6	11.8	11.2					<0.001	0.480

[1]SEM for gender × Lys.

[2]Lin. = Linear; Quad. = Quadratic.

[3]G = gender.

[4]Lys × G = interaction for Lys × gender.

1.0% Lys) were fed for 5 wk until slaughter at an average of 130 kg BW. Boler et al. (2011b) found that lean yields were increased as the concentration of dietary Lys increased from 0.7 to 0.9% Lys and concluded that IC males should be fed increased concentrations of Lys compared with physical castrates. However, the authors did not investigate how these Lys requirements may change over time as the potential of the IC male pig to deposit lean tissue changes from being similar to that of an entire male to be more similar to that of a castrate.

The NRC (2012) swine growth model was used to predict SID Lys requirements based on the BW, feed ME, and ADFI associated with the time periods (d 0–14 and d 15–28) following the second immunization against GnRF in this experiment. For the 2-wk period following the second immunization against GnRF (60.1 to 74.7 kg BW), the NRC model predicts

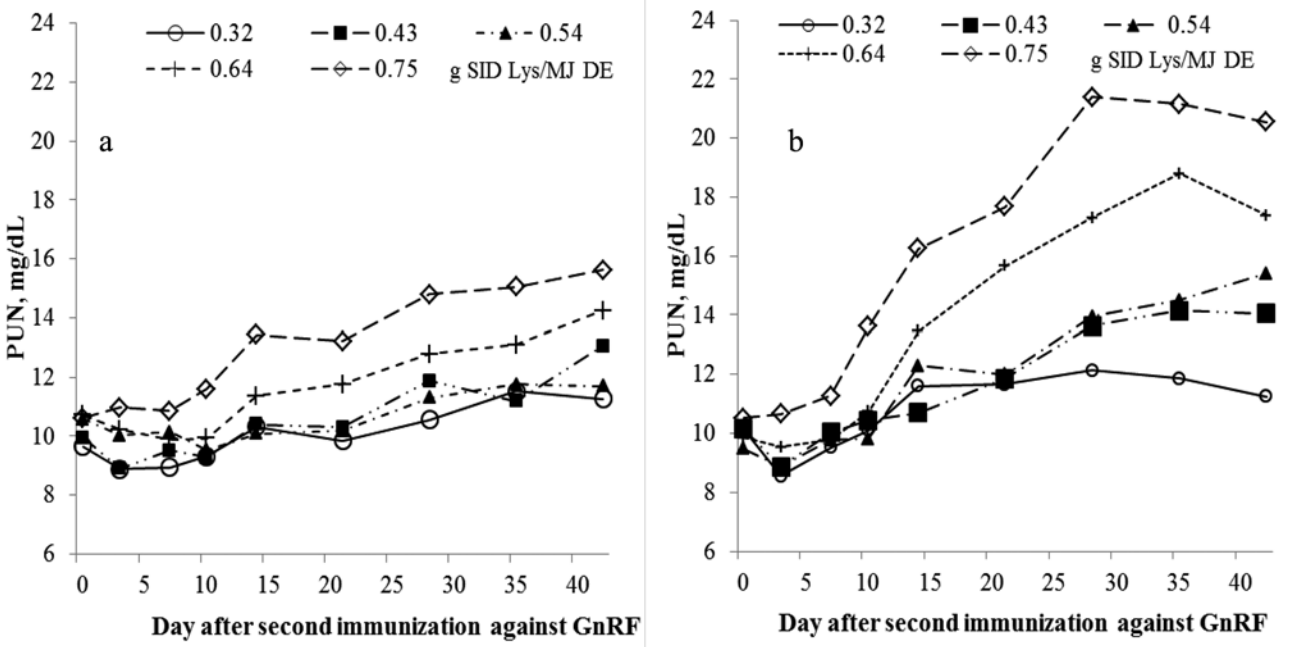

Figure 4. Change in plasma urea nitrogen (PUN) concentration over 6 wk for (a) entire males and (b) immunocastrated males from 60.1 to 107 kg BW fed increasing concentrations of available Lys (n = 21). GnRF = gonadotrophin releasing factor; SID = standardized ileal digestible.

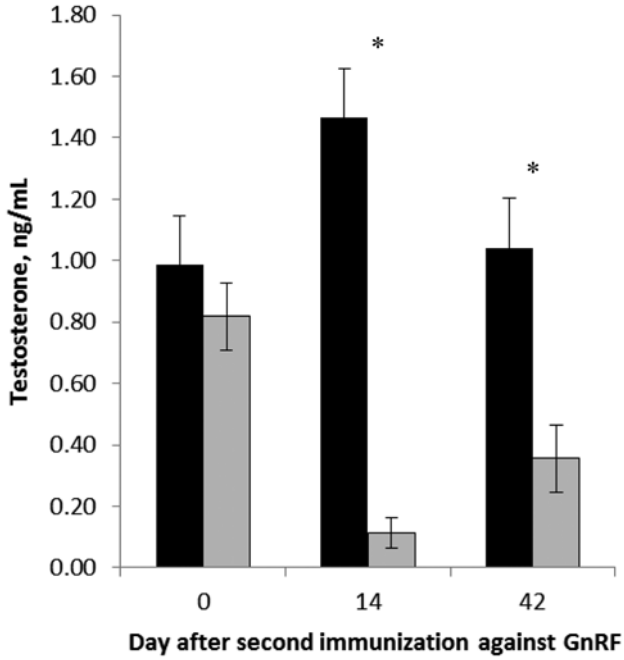

Figure 5. Testosterone concentration on d 0, 14, and 42 after the second immunization against gonadotrophin releasing factor (GnRF) for entire males (black) and immunocastrated males (shaded; n = 30). The P-values for gender for d 0, 14, and 42 were $P = 0.301$, $P < 0.001$, and $P < 0.001$, respectively. Data for each Lys concentration has been pooled due to a lack of treatment effect ($P > 0.05$).

Table 4. Predicted Lys requirements (g standardized ileal digestible Lys/MJ DE) for entire male and immunocastrated (IC) male pigs from 60.1 to 107.5 kg BW using a linear plateau model or a quadratic polynomial model

		Linear plateau		Quadratic polynomial	
Item	Gender	Requirement[1]	R^2	Requirement	R^2
d 0–14					
ADG	Entire male	0.68	0.94	0.83	0.97
	IC male	0.64	0.86	0.69	0.85
G:F	Entire male	0.72	0.95	–[2]	0.97
	IC male	0.64	0.92	0.81	0.90
d 15–28					
ADG	Entire male	0.62	0.96	–	0.99
	IC male	0.43	0.66	–	0.40
G:F	Entire male	0.60	0.91	–	–
	IC male	0.43	0.34	–	–
d 29–42					
ADG	Entire male	0.54	0.93	0.62	0.92
	IC male	0.38	0.43	0.54	0.72
G:F	Entire male	0.54	0.78	0.59	0.87
	IC male	0.36	0.06	0.51	0.93
d 0–42					
ADG	Entire male	0.58	0.99	0.71	1.00
	IC male	0.40	0.91	0.64	0.75
G:F	Entire male	0.51	0.99	0.68	0.98
	IC male	0.42	1.00	0.61	0.86

[1]Predicted Lys response was calculated using formulated dietary Lys values.

[2]Unable to solve with this method within the range of Lys values.

the average requirement to be 0.52 g SID Lys/MJ DE, whereas for second 2-wk period following the second immunization against GnRF (74.7 to 92.8 kg BW), it is predicted to be 0.48 g SID Lys/MJ DE. In comparison, the findings from the current experiment suggest the requirements for IC males are approximately 0.65 and 0.43 g SID Lys/MJ DE for d 0 to 14 and d 15 to 28, respectively (where d 0 is the second immunization against GnRF). This suggests that the initial requirements of IC pigs are higher than predicted by the NRC growth model and decrease more quickly than is perhaps being predicted in the NRC growth model following the second immunization against GnRF.

Plasma urea nitrogen concentrations in IC males were found to increase from d 10 after the second immunization against GnRF. An increase in the PUN concentration indicates that there are excess AA that were deaminated and converted to urea nitrogen (Dunshea et al., 1992, 2013). As indicated earlier, this provides further support to the findings in growth performance that the SID Lys concentrations in the diet of IC males can be decreased beyond 2 wk after the second immunization against GnRF. Dunshea et al. (2013) also arrived to a similar conclusion after examining hormonal, metabolic, and feed intake responses after the second immunization against GnRF.

In a nitrogen retention experiment, Huber et al. (2013) found that by approximately d 8 after the second immunization against GnRF, the PUN concentration tended to be less in entire males compared with IC males with the PUN concentrations of IC males approaching those of physical castrates between d 7 and 16 following the second immunization against GnRF. In agreement with the current experiment, Bauer et al. (2009) also found that PUN concentrations began to increase within 10 d after the second immunization against GnRF whereas Claus et al. (2007) found that the PUN concentration increased immediately after the second immunization against GnRF. Huber et al. (2013) attributes the differences in changing PUN concentrations following the second immunization between studies to variation in feed intake and protein deposition patterns.

The increased PUN concentrations of IC males can also be partly attributed to the increase in feed intake associated with IC males after the second immunization (Dunshea et al., 2013). In addition, the increased PUN concentration also shows that IC males have elevated nitrogen excretion compared with entire males. Decreased nitrogen retention occurs due to the

decrease in anabolic hormones in IC male pigs (Claus et al., 2007).

The response to Lys in this experiment was estimated by the linear-plateau model and a quadratic model. The choice of model to analyze the data can give different results (Baker, 1986; Dunshea et al., 2000). Baker (1986) stated that the broken-line response describes the requirements of the average animal in a population. It also assumes that an individual animal responds linearly to additions of, for example, Lys until the requirement is met and that after this there will be no further improvement (Coma et al., 1995). In contrast, the curvilinear or quadratic method provides the requirements for a maximum response from all the animals in the population. Therefore, the quadratic method can overestimate the requirements whereas the linear-plateau model may underestimate the requirements depending on the data set (Williams et al., 1984; Robbins et al., 2006).

The increase in ADFI, ADG, G:F, HCW, and backfat by the IC males after the second immunization against GnRF is consistent with findings of previous studies (Dunshea et al., 2013; Lanferdini et al., 2013). A meta-analysis by Dunshea et al. (2013) found that IC males showed an increase in daily gain (+199 g/d), an increase in feed intake (+429 g/d), an increase in feed conversion ratio (+0.11), an increase in HCW (+2.09 kg), and an increase in backfat (+1.53 mm) compared with entire males. This concurs with the findings by Batorek et al. (2012), who also conducted a meta-analysis but with studies that used both Improvac and experimental vaccines. The increase in feed intake by IC males is due to both the effects of the change in sex hormones on metabolism and a decrease in aggression and sexual behavior due to the absence of estrogens and androgens, and therefore, more time is spent eating (Claus, 1987 [as cited in Claus et al., 2007]; Dunshea et al., 2001; Cronin et al., 2003).

Dressing percentage was decreased by 0.7% in IC males compared with entire males. A meta-analysis of 11 studies by Dunshea et al. (2013) found that dressing percentage was decreased by 0.3% across these studies, which is lower than in the present experiment. Boler et al. (2014) found that IC males had an increased gut fill, liver size, and intestinal mass compared with entire males, which may help to explain the decreased dressing percentage observed in this experiment.

Conclusions

Immunocastrated male pigs should be fed SID Lys concentrations similar to those of entire male pigs for 2 wk after the second immunization. Beyond this, the SID Lys concentration in the diet should be reduced for IC males. In the BW range used in this experiment, we recommend that IC males are fed diets containing approximately 0.65 g SID Lys/MJ DE for the first 2 wk after the second immunization against GnRF. The dietary SID Lys concentration can then be decreased to approximately 0.43 g SID Lys/MJ DE. This ensures the producer can capitalize on the production benefit of the entire male and then can decrease Lys concentrations in the diet to reduce costs and optimize returns as the pig becomes more similar to a physical castrate.

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