## Dietary nitrogen content affects lysine requirement and nitrogen utilization and retention in growing pigs

Miranda Buchinski<sup>1,2,</sup> Carley Camire<sup>1,2,</sup> Kate Shoveller<sup>3,</sup> Dan Columbus<sup>1,2,</sup>

- <sup>1</sup>Prairie Swine Centre, Inc.
- <sup>2</sup> Department of Animal and Poultry Science, University of Saskatchewan
- <sup>3</sup> Animal Biosciences, University of Guelph

Reducing protein content in diets while supplementing crystalline amino acids to meet essential amino acid requirements has become commonplace. In general, these diets have been successful at maintaining growth performance while reducing protein (i.e., nitrogen) waste into the environment. However, in some situations, for

example when dietary protein is reduced by more than 3%, non-essential amino acids or total dietary protein may become limiting for maximum nitrogen retention (i.e., lean gain) and growth performance. For example, Guay et al. (2006) and Jansman et al. (2016) observed reduced growth performance in pigs fed low protein diets even though a sufficient amount of essential AA had been added. This suggests that non-essential amino acids may become essential when dietary protein is below a critical level. Nitrogen deficiency may limit essential amino acid utilization and result in changes in essential amino acid requirements.

It has been suggested that the ratio of essential amino acid-nitrogen to total nitrogen ratio (E:T) can be used as an indicator of nitrogen sufficiency in the diets. At extreme ratios, nitrogen utilization suffers due to a lack of essential amino acids (i.e., low ratios) or non-essential amino acids/nitrogen (i.e., high ratios). Heger et al. (1998) estimated an optimum ratio of 0.48 in pigs for maximum nitrogen retention, however, this ratio was calculated using total essential amino acids and only amino acid nitrogen. Advances in our understanding of nitrogen metabolism have shown that pigs are capable of utilizing sources of non-protein nitrogen (NPN; e.g., urea, ammonia) to meet amino acid requirements, especially for non-essential amino acids. Therefore, we have suggested that the E:T ratio should be calculated as the standardized ileal digestible (SID) essential amino acid nitrogen up to requirements (E) and nitrogen from all other sources (T; crude protein).

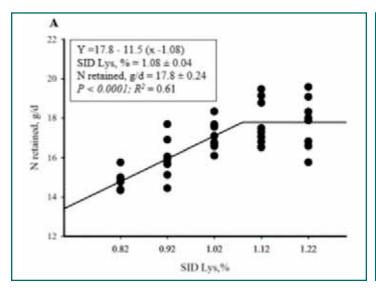
Our objectives were to:

- 1. Determine the effect of E:T ratio on the lysine requirement for nitrogen retention in growing pigs.
- 2. Determine the effect of E:T ratio and inclusion of non-protein nitrogen (i.e., ammonium phosphate) on the lysine requirement for nitrogen retention in growing pigs.
- 3. Determine the effect of E:T ratio, lysine content, and non-protein nitrogen inclusion on growth performance, nitrogen output, and body composition of growing pigs.

## Methodology

Study 1 (Objective 1): A total of 80 growing barrows with an initial body weight of  $21.5 \pm 0.89$  kg were randomly assigned to 1 of 10 diets (n = 8) in 8 blocks in a 2 × 5 factorial arrangement. Diets consisted of a low E:T ratio (LR; 0.33) or a high E:T ratio (HR; 0.36) with graded lysine content (0.8%, 0.9%, 1.0%, 1.1%, and 1.2% SID) fed at  $2.8 \times \text{maintenance}$  metabolizable energy requirements in 2 equal meals each day. The E:T ratio was adjusted in these diets by altering the soybean meal content in the diet while keeping the essential amino acid content constant. After a 7-d adaptation, a 4-d nitrogen-balance collection was conducted and nitrogen retention was calculated as the difference between nitrogen intake (diet) and output (urine and feces).

Study 2 (Objective 2): A total of 90 growing barrows with an initial body weight of  $20.4 \pm 0.46$  kg were randomly assigned to 1 of 10 dietary treatments (n = 9 pigs/treatment) in 9 blocks in a  $2 \times 5$  factorial design. Diets contained no ammonium phosphate (NAP; E:T of 0.36) or were supplemented with 1.7% ammonium phosphate (AP; E:T of 0.33) as a source of NPN with graded levels of dietary lysine [0.8%, 0.9%, 1.0%, 1.1% and 1.2% SID] fed at  $2.8 \times$  maintenance metabolizable energy requirements in 2 equal meals each day. The E:T ratio was adjusted in these diets by altering the inclusion of ammonium phosphate while keeping the essential amino acid content constant. After a 7-d adaptation, a 4-d nitrogen-balance collection was conducted and nitrogen retention was calculated as the difference between nitrogen intake (diet) and output (urine and feces).



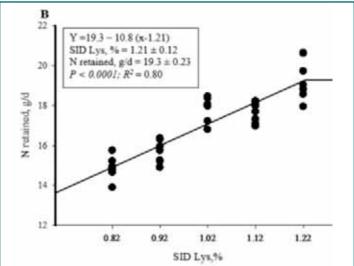
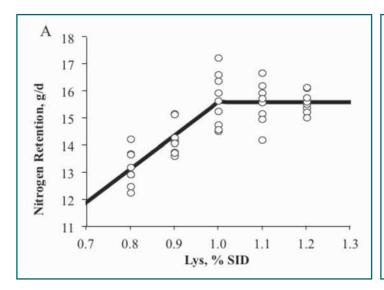


Figure 1. The linear broken-line model estimated nitrogen retention (N retention; g/d) in pigs fed high (HR; 0.36) or low (LR; 0.33) E:T ratio diet. A breakpoint was achieved at 1.08 SID Lys, % with a maximum N retention of 17.8 g/d for pigs fed the HR diets (Fig. 1A). While the breakpoint was achieved in pigs fed the LR diet at 1.21 SID Lys, % with a maximum N retention of 19.3 g/d (Fig.1B).



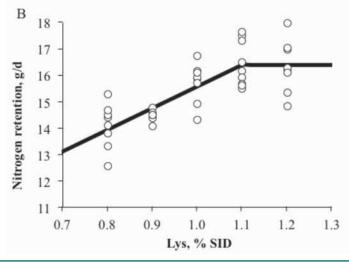


Figure 2. The two-phase breakpoint analyses estimates for nitrogen retention (NR; g/d) in pigs fed no ammonium phosphate (NAP) and ammonium phosphate (AP). The analyses indicated a breakpoint of 1.00% with maximum NR at 15.6 g/d in pigs fed the NAP diet (A). A breakpoint of 1.09% with maximum NR at 16.4 g/d was achieved in pigs fed the AP diet (B).

Study 3 (Objective 3): A total of 240 mixed-sex growing pigs with an initial body weight of 20.2 ± 2.18 kg were housed in groups of 5 pigs/pen. Pens were randomly assigned to 1 of 6 dietary treatments over 3 blocks (n = 8 pens/treatment) in a 2 × 3 factorial design, with factors of NPN inclusion (no ammonium phosphate [NAP] or ammonium phosphate inclusion at 1.7% [AP]) and dietary lysine (1.03%, 1.15% or 1.27% SID). The NAP and AP diets were formulated to have an E:T ratio of 0.35 and 0.33, respectively. Pigs had ad libitum access to feed and water for the duration of the experiment (28 d). Individual pig body weight and feed intake were measured weekly to determine average daily gain (ADG), average daily feed intake (ADFI), and gain:feed (G:F). Fresh fecal samples were obtained on d 15 to determine digestibility. On d 28, backfat and lean depth were measured on 2 pigs per pen via ultrasound.

## Results

Study 1 (Objective 1): There was a significant interaction between E:T ratio and, where LR diets had a higher nitrogen retention than HR diets, while increasing lysine linearly increased nitrogen retention in both HR and LR diets. The marginal efficiency of utilizing SID lysine reduced with increasing lysine content, while the efficiency of utilizing N increased as lysine increased. The SID lysine required to maximize nitrogen retention of HR-fed pigs was estimated at 1.08% (R2 = 0.61) and at 1.21% (R2 = 0.80) in LR-fed pigs (Figure 1).

**Study 2 (Objective 2):** Lysine and nitrogen content had an effect on fecal and urinary nitrogen output, including a decrease (Dietary nitrogen content ... cont'd on page 10)

(Dietary nitrogen content... cont'd from page 9)

in urinary nitrogen and an increase in nitrogen retention with inclusion of NPN and increasing lysine (P < 0.01). The marginal efficiency of nitrogen was improved with increasing lysine content, but reduced with inclusion of NPN. Marginal efficiency of lysine was decreased with increasing lysine content, but improved with NPN inclusion. The linear breakpoint model indicated NR was maximized at 1.00% SID lysine (15.6 g/d NR; R2 = 0.68) in NAP-fed pigs and at 1.09% SID lysine (16.4 g/d NR; R2 = 0.61) in AP-fed pigs (Figure 2).

Study 3 (Objective 3): Overall ADG and d 28 body weight increased with increasing lysine, but were not impacted by dietary NPN content (Table 1). Inclusion of NPN reduced feed intake and increased G:F compared to pigs fed NAP diets. Inclusion of NPN increased fecal N output. Pigs fed AP diets had increased lean depth with no effect on backfat (Table 2).

## Summary

These results indicate that:

- Non-essential amino acids, or total dietary nitrogen, become limiting in diets with a high E:T ratio. This deficiency can be mitigated through supplementation with either intact protein or a source of non-protein nitrogen (i.e., ammonium phosphate)
- An increase in dietary lysine is required when diets contain sufficient nitrogen as a result of improved nitrogen retention (i.e., lean gain)
- Including a source of non-protein nitrogen improved feed efficiency while maintaining growth performance, indicating that ammonium phosphate is an appropriate source of nitrogen in swine diets.
- Nutritionists should consider the E:T in diet formulation as an indication of N sufficiency.

Table 1. Growth performance metrics for diets with and without ammonium phosphate at 1.7% inclusion with increasing SID Lys content<sup>1</sup>

	No ammonium phospha			Amm	onium pho	osphate		Р	'-valuesDor	esDom	
Lys, % SID	1.03	1.15	1.27	1.03	1.15	1.27	SEM	N	Lys	N × Lys	
Body Weight, kg									·		
Day 0	20.2	20.2	20.4	20.3	19.9	20.2	0.38	0.30	0.32	0.36	
Day 7	25.1	25.5	25.4	25.0	24.8	25.2	0.37	0.08	0.44	0.36	
Day 14	31.1	31.7	31.6	30.8	30.8	31.8	0.35	0.17	0.05	0.14	
Day 21	38.1	39.1	39.1	37.4	37.7	39.3	0.47	0.02	<0.001	0.06	
Day 28	45.3	46.7	46.5	44.9	45.1	46.9	0.65	0.19	0.01	0.10	
Average daily gain, kg/d											
Days 0-7	0.70	0.73	0.72	0.69	0.70	0.72	0.016	0.31	0.38	0.86	
Days 8-14	0.88	0.90	0.88	0.82	0.85	0.95	0.031	0.62	0.17	0.16	
Days 15-21	0.99	1.06	1.07	0.95	0.99	1.02	0.032	0.02	0.03	0.83	
Days 22-28	1.03	1.08	1.07	1.06	1.05	1.10	0.038	0.74	0.65	0.73	
Days 0-28	0.90	0.93	0.94	0.88	0.90	0.94	0.017	0.24	0.02	0.73	
Average daily feed intake, kg/d											
Days 0-7	0.70	0.73	0.72	0.69	0.70	0.72	0.017	< 0.001	0.11	0.30	
Days 8-14	0.88	0.90	0.88	0.82	0.85	0.95	0.023	0.003	0.19	0.30	
Days 15-21	0.99	1.06	1.07	0.95	0.99	1.02	0.039	0.01	0.14	0.77	
Days 22-28	1.03	1.08	1.07	1.06	1.05	1.10	0.064	0.01	0.51	0.37	
Days 0-28	0.90	0.93	0.94	0.88	0.90	0.94	0.027	< 0.001	0.27	0.51	
Gain:Feed, kg/kg											
Days 0-7	0.60	0.60	0.59	0.61	0.63	0.62	0.010	0.01	0.73	0.60	
Days 8-14	0.57	0.60	0.60	0.60	0.60	0.65	0.013	0.02	0.02	0.28	
Days 15-21	0.60	0.63	0.62	0.60	0.62	0.62	0.014	0.85	0.32	0.97	
Days 22-28	0.57	0.57	0.54	0.57	0.59	0.58	0.016	0.14	0.61	0.47	
Days 0-28	0.58	0.59	0.59	0.59	0.61	0.61	0.006	0.002	0.08	0.51	

ADFI, average daily feed intake; ADG, average daily gain; G:F, gain:feed; Lys, lysine; N, nitrogen; SID, standardized ileal digestible; SEM, standard error of the mean.

<sup>&</sup>lt;sup>1</sup>Data presented are least-square means (n=8 pens/treatment).

Table 2. Backfat and lean depth measurements from pigs fed diets not including or including ammonium phosphate at 1.7% inclusion with increasing SID Lys content<sup>1</sup>

	No ammonium phosphate			Ammonium phosphate			P-values			
Lys, % SID	1.03	1.15	1.27	1.03	1.15	1.27	SEM	N	Lys	N×L
Backfat, mm	6.6	6.7	6.8	6.4	6.4	6.5	0.32	0.15	0.80	0.89
Lean, mm	36.5	37.9	37.3	38.0	40.1	38.9	1.05	0.02	0.13	0.91

Lys, lysine; N, nitrogen; SID, standardized ileal digestible; SEM, standard error of the mean.



<sup>&</sup>lt;sup>1</sup>Data presented are least-square means (n=8 pens/treatment).