Arginine supplementation for pigs weaned at 21 days of age

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Executive Summary

Young mammals including pigs have a high dietary requirement for arginine, predominantly due to its role as a nitrogen carrier in tissue proteins, and its key role in gastrointestinal growth and development. Hernandez *et al.* (2009) showed that the addition of 0.6% arginine to the diets of 21-day-old newly-weaned pigs, which weighed less than 5 kg, for 10 days after weaning significantly improved daily feed intake and daily gain in the dietary transition period between days 11 and 14 post-weaning. We tested the hypothesis that supplementing arginine at different rates ranging from 0 to 1.0% would increase the growth performance and survivability of newly weaned, 21 day-old pigs.

One thousand two hundred entire male pigs (Large White x Landrace-PrimeGro™ Genetics) were individually weighed and selected at weaning (21 days of age) and allocated to pens of 10 pigs of similar weaning weight. In a 2 x 5 factorial design, pens containing heavy (greater than 6 kg) or light (less than 5.5 kg) piglets were allocated to one of five diets containing 0%, 0.25%, 0.5%, 0.75% or 1.0% supplemental synthetic arginine. Pigs were offered their respective arginine diets for 12 days post-weaning. The average weaning weights for the light and heavy treatments were 4.84± 0.02 kg and 7.00± 0.09 kg, respectively. The diets were formulated to contain 15.2 MJ digestible energy/kg and 0.93 g av. lysine/MJ DE. Common commercial weaner diets were fed to all pigs from day 13 post-weaning to day 40 (end of the weaner phase). The pigs were weighed at weaning, 12, 20 and 40 days post-weaning. The feed intake for the pigs was also recorded at these times. The experimental unit for all analyses was the pen of animals. The data were analysed by General Linear Model, ANOVA and linear or quadratic relationships were also analysed.

There were no significant dose dependent responses to supplemental arginine in either heavy or light pigs during the weaner period. Over the first 12 days during supplementation there was a trend for the heavier pigs to grow slower than the light pigs (0.101g/d vs. 0.114 g/day, respectively). Over the whole weaner period, pigs that were lighter at weaning had a significantly lower feed intake and average daily gain. There were no significant improvements (P>0.05) in rate of gain, daily feed intake and feed conversion rate of light and heavy classes of pig fed arginine for 12 days post–weaning. Furthermore, there were no significant differences in mortality or removals (unthrifty pigs) between the treatments for the period between 0 and 40 days post weaning (light weight class, P>0.05; x^2=1.49; heavy weight class, P>0.05; x^2=4.41).

It was concluded that pigs weaned at 21 days under commercial conditions and offered weaner diets that contain a mixture of grains, vegetable proteins and animal proteins do not require supplemental dietary arginine and that these results support those of others, in that endogenous supply of arginine through *de-novo* biosynthesis is adequate using commercial dietary ingredients.

Table of Contents

1. Introduction

Young mammals including pigs have a high dietary requirement for arginine predominantly due to its role as a nitrogen carrier in tissue proteins and its key role in gastrointestinal growth and development. As an amino acid, arginine is metabolized into glutamate for entry to the oxidative reactions of the citric acid cycle to yield energy, whilst the nitrogen atoms are hydrolysed to form urea. Some researchers consider arginine to be a key essential amino acid for maximal growth of the young pig (Wu *et al*., 2007), especially since piglet plasma concentrations decline throughout a sow's lactation (Wu *et al.,* 2004; 2007; Flynn *et al*., 2000). Kim *et al.* (2004) reported improvements in growth rate commensurate with increased plasma arginine, insulin and growth hormone levels when 7-day-old weaned piglets were offered synthetic arginine in liquid-based milk diets. These data suggest that sub-optimal arginine intake during suckling could place the young pig in a state of arginine deficiency when it is weaned. On the other hand, Ball *et al*. (2007) argued that dietary intake and *de novo* synthesis of arginine from dietary protein sources should be sufficient to meet the metabolic arginine requirement in young piglets. The National Research Council (NRC, 1998) recommends a requirement for total dietary arginine for pigs weighing 3 to 5 kg of 0.59% and in pigs weighing 5 to 10 kg a requirement of 0.54%.

Whilst research has been directed at increasing arginine levels in piglets in the pre-weaning period, little or no research has been conducted examining what occurs in the post-weaning period where newly weaned pigs have to endure a myriad of changes. These changes cause a post-weaning "growth check" that is characterised by damage to mucosal epithelium of the gastrointestinal tract that in turn, can compromise growth performance (Pluske *et al*., 1997). Damage to the mucosal epithelium reduces metabolism of essential amino acids such as arginine, and hence provision of synthetic amino acids in the diet offers a means of maintaining and re-building the intestinal epithelium and reducing the growth check. As part of the CRC project 2B-103, Hernandez *et al.* (2009) investigated the addition of 0 vs. 0.6% arginine to the diets of newly-weaned, 21 day old pigs, which weighed less than 5 kg, for 10 days after weaning. They did not show any improvement in growth performance or feed intake during the ten day supplementation period. However, surprisingly they found significant improvements in daily feed intake and average daily gain between days 11 to 14 post-weaning, after the arginine supplementation had ceased. Over the whole 21 day weaner period this improvement in growth performance and feed intake caused a tendency for the arginine supplemented pigs to eat more feed (10%) and grow faster (15%). The authors had no obvious explanation for this finding, however they speculated that there may have been carry-over effects on subsequent metabolism between days 10 and 14. Furthermore the arginine supplementation may have improved gastrointestinal tract development and there may have been increased small intestine absorption due to an increase in villous height and microvascular development which could have positively affected growth performance and feed intake. Hernandez *et al.* (2009) concluded that it was necessary to investigate on a larger scale, arginine supplementation in dry diets. Therefore, as a continuation of that project (Pork CRC project 2B-103), we investigated the effect of providing additional arginine to the diets of newly weaned, 27 day old pigs (Morrison *et al*. 2009). We found there was no benefit of supplementing 27 day old weaned pigs with arginine for 12 days post-weaning. However, we calculated that given the intake of these pigs post-weaning, the pigs offered no supplemental arginine (control treatment) would have met the daily intake requirements of 0.54 g arginine/100g intake as recommend by the NRC, and were therefore not limited in their arginine intake.

Under commercial conditions, low feed intakes could result in inadequate arginine intake. Therefore it was essential to conduct further research to investigate the benefits of feeding supplemental arginine to weaned pigs at a younger age and in piglets with a low weaning weight, and hence low appetite. The aim of this experiment was to investigate the effect of arginine supplementation ranging from 0 to 1% on the growth performance of 21 day old weaned pigs that were classified as light or heavy weight at weaning. The hypothesis was that weaned pigs supplemented with 1% arginine would have increased growth performance and survivability in the post-weaning period, and that the response to arginine is greater in light-weaned piglets.

2. Methodology

The Rivalea Animal Care and Ethics Committee approved this experiment (09N051C/2009). One thousand two hundred male pigs (Large White x Landrace-PrimeGro™ Genetics) were selected at weaning (21 days of age) and allocated to pens of 10 pigs based on individual weaning weight (light weaning weight: pigs below 5.5 kg; and heavy weaning weight: above 6.0 kg). Pens were allocated to a 2 x 5 factorial experiment with the respective factors being weaning weight (light and heavy) and arginine supplementation (0, 0.25%, 0.5%, 0.75% and 1.0% arginine). The pigs were selected over a 12 week period (100 pigs per week) commencing November 2009. The average weights (mean±SE) at weaning for the light and heavy weaners were 4.84± 0.02 kg and 7.00± 0.09 kg, respectively.

The treatments with the level of supplemental arginine were as follows:

A. Heavy weaners -0% arginine
B. Heavy weaners - 0.25% arginine
C. Heavy weaners - 0.5% arginine
D. Heavy weaners - 0.75% arginine
E. Heavy weaners -1% arginine
F. Light weaners - 0% arginine
G. Light weaners - 0.25% arginine
H. Light weaners - 0.5% arginine
I. Light weaners - 0.75% arginine
J. Light weaners - 1% arginine

The arginine used was L-Arginine Monohydrochloride (supplied by Optigen Ingredients Pty Ltd.). The treatment diets were fed for 12 days post-weaning (in the starter diet). From day 13 to 42 days post-weaning, the pigs were fed commercial weaner diets (phase 1 and phase 2) that were not supplemented with additional arginine.

The following measurements were made: Pen weaning weights at day 0, 12 (end of starter diet), 20 and 42 days post-weaning. The pen feed intake was recorded, and FCR calculated between 0-12 days post-weaning, 12-20 days post-weaning and 20-42 days post-weaning. The number of deaths and removals post-weaning were also recorded. The data were analysed by General Linear Model analysis of variance and analysis of linear or quadratic relationships were conducted using SPSS Statistical version 17 software (2007). The mortality data were analysed by chi-squared analysis.

Table 1 - Ingredient composition and nutrient profile of the starter weaner diets offered during the first 12 days post-weaning, % of diet (as fed basis).

Ingredient	%
Wheat	20.00
Groats	24.74
Soybean meal	6.5
Nupro Alltech	4.2
Meatmeal	3.20
Fishmeal	6.20
Bloodmeal	2.50
Whey powder	25.00
Water	1.00
Natuphos 5000	0.01
Tallow-enzyme	2.00
Tallow-mixer	2.3
Salt	0.20
Lysine HCL	0.35
Dl-methionine	0.13
Threonine	0.17
Isoleucine	0.10
Tryptophan	0.07
Zinc oxide	0.28
QAF creep premix	0.30
CTC	0.20
Pulmotil 200 premix	0.20
Acid Lac dry	0.3
Ronozyme	0.03
Lysoforte	0.067
Adimix 30% coated	0.3

*Estimated from composition of ingredients (SCA 1987).

Table 2 - Nutrient profile of each of the commercial weaner diets, % of diet (as fed basis)

Nutrient composition, %*	*Base Starter*	*Phase 1 Weaner*[1]	*Phase 2 Weaner*[1]
DE, MJ/kg	15.32	14.74	14.50
Crude protein	22.90	24.01	23.52
Crude fat	7.28	5.38	4.78
Crude fibre	1.69	2.49	2.85
Total Lysine	1.63	1.54	1.40
Available lysine	1.47	1.37	1.22

*Estimated from composition of ingredients (SCA 1987). [1]Diets not further supplemented with arginine.

3. Outcomes

Table 3 summarises the average daily gain, average daily feed intake, feed conversion rate of light and heavy weaner pigs fed diets supplemented with 0 to 1.0% arginine for 12 days post-weaning. There was no significant impact ($P>0.05$) of dietary arginine supplementation on the average daily gain, average daily feed intake or feed conversion ratio from weaning to 12 days post-weaning, from 12 to 20 days post-weaning, from 0 to 20 days post-weaning, or overall, from 0 to 40 days post-weaning. There were no significant quadratic or linear relationships observed ($P>0.05$) between supplemental arginine as a main effect or as an interaction with weaning weight.

The weight class of the pig at weaning had a significant impact on growth performance factors measured in the post-weaning period. There was a trend ($p=0.092$) for lighter pigs to grow faster than heavier pigs (0.114±0.004 kg/day vs. 0.101±0.004 kg/day, respectively) in the 12 days post-weaning. Lighter pigs consumed significantly less ($P<0.05$) food than the heavier pigs (0.143 ±0.003 kg/day vs. 0.157±0.004 kg/day, respectively) in the 12 days post-weaning. As a result of this, the feed efficiency of the lighter pigs was significantly lower ($P<0.001$) in the lighter pigs compared to the heavier pigs (1.35±0.03 vs. 1.69±0.05, respectively). For the time period between 12 to 20 days, 20 to 40 and 0 to 40 days post-weaning (overall) the lighter pigs consumed significantly ($P<0.05$) less food and grew slower than their heavier counterparts. There was no significant ($P>0.05$) difference in feed efficiency during these time periods.

There were no significant differences in mortality or removals (unthrifty pigs) between the treatments for the period between 0 and 40 days post-weaning. The number of deaths and removals were 17/120, 11/120, 12/120, 12/120, and 20/120 out of 0%, 0.25%, 0.5%, 0.75% and 1.0% arginine treatments, respectively in the light weight class ($P>0.05$; $\chi^2=1.49$). In the heavy weight class, there were also no significant differences in mortality removals (unthrifty pigs) between the treatments for the period between 0 and 40 days post weaning ($P>0.05$; $\chi^2=4.41$). The number of deaths were 6/120, 8/120, 7/120, 2/120 and 3/120 out of each 0%, 0.25%, 0.5%, 0.75% and 1.0% arginine treatments, respectively. Regardless of treatment, there was a difference in the number of deaths and removals between the light and heavy weight classes of pigs. There were significantly ($P<0.001$; $\chi^2=23.51$) more deaths and removals from the lighter class of pig at weaning.

Table 4 summarises the arginine percentage of the diets and arginine intakes of the pigs in light and heavy weight classes.

Table 3 - The influence of 0 to 1.0% arginine supplementation on growth performance of light (below 5.5kg) and heavy (greater than 6 kg) pigs at weaning in the post-weaning period.

	Light					Heavy					SEM	Significance		
	0% Arg	0.25% Arg	0.5% Arg	0.75% Arg	1.0% Arg	0% Arg	0.25% Arg	0.5% Arg	0.75% Arg	1.0% Arg		Diet	Weaning Weight	Diet x Wean. weight
Av weight weaning (kg)	4.83	4.83	4.85	4.86	4.83	7.01	7.01	6.99	7.07	6.93	0.101	0.808	<0.001	0.947
Av weight day 40 (kg)	19.69	20.07	19.39	19.85	19.83	23.55	24.03	23.65	23.67	24.42	0.227	0.512	<0.001	0.872
Av. daily gain (kg/d)														
Weaning to day 12	0.109	0.135	0.106	0.107	0.115	0.084	0.102	0.105	0.110	0.104	0.004	0.540	0.092	0.570
Day 12 to day 20	0.349	0.326	0.321	0.357	0.316	0.351	0.386	0.382	0.371	0.386	0.007	0.967	0.003	0.389
Day 20 to day 40	0.534	0.547	0.528	0.540	0.557	0.633	0.625	0.624	0.614	0.652	0.006	0.373	<0.001	0.889
Weaning to day 40	0.371	0.380	0.361	0.374	0.377	0.413	0.422	0.421	0.414	0.435	0.004	0.464	<0.001	0.724
Av. daily intake (kg/d)														
Weaning to day 12	0.148	0.153	0.135	0.140	0.141	0.143	0.165	0.149	0.169	0.160	0.003	0.205	0.007	0.267
Day 12 to day 20	0.365	0.385	0.334	0.380	0.342	0.362	0.365	0.381	0.431	0.417	0.009	0.480	0.099	0.434
Day 20 to day 40	0.749	0.829	0.710	0.743	0.809	0.863	0.802	0.775	0.842	0.899	0.014	0.161	0.015	0.519
Weaning to day 40	0.478	0.521	0.451	0.476	0.500	0.532	0.517	0.503	0.555	0.578	0.007	0.081	<0.001	0.328
Feed conversion rate (g/g)														
Weaning to day 12	1.38	1.24	1.28	1.41	1.44	1.94	1.67	1.55	1.65	1.63	0.044	0.351	0.00	0.558
Day 12 to day 20	1.06	1.20	1.08	1.09	1.20	1.08	0.94	1.02	1.18	1.11	0.031	0.808	0.368	0.465
Day 20 to day 40	1.42	1.54	1.37	1.38	1.48	1.38	1.29	1.25	1.38	1.38	0.028	0.742	0.087	0.732
Weaning to day 40	1.3	1.38	1.27	1.27	1.34	1.30	1.23	1.20	1.34	1.34	0.019	0.596	0.456	0.475

Table 4 - Arginine % in diets and average daily arginine intake of light and heavy pigs between weaning and 12 days post-weaning

	Light					Heavy				
	0% Arg	0.25% Arg	0.5% Arg	0.75% Arg	1.0% Arg	0% Arg	0.25% Arg	0.5% Arg	0.75% Arg	1.0% Arg
Total dietary arginine (%)	1.10	1.34	1.58	1.82	2.05	1.10	1.34	1.58	1.82	2.05
Av. daily feed intake (kg/day)	0.148	0.153	0.135	0.140	0.141	0.143	0.165	0.149	0.169	0.160
Av. daily arginine intake between weaning and day 12 (g/day)	1.63	2.05	2.13	2.55	2.89	1.57	2.21	2.35	3.04	3.36

4. Application of Research/Discussion

The hypothesis was rejected in the current experiment. The results have shown that there was no benefit of providing supplemental synthetic arginine to the commercial diets of newly weaned, 21 day old pigs in either light or heavy weight classes. There were no significant growth performance improvements or improvements in survivability of pigs fed supplemental arginine at 0, 0.25, 0.5, 0.75 or 1.0% for 12 days post-weaning.

It is well accepted that arginine is an essential amino acid in pigs, and there may be an additional dietary requirement in the neonate (Wu *et al*., 2007), but in the healthy adult, endogenous synthesis is considered adequate to meet the pigs' requirement (Easter *et al*., 1974). The metabolic requirements for arginine in young pigs are influenced mainly by demand for protein deposition, and it has been estimated that approximately 70% of the daily arginine used is for protein synthesis (Wu *et al*., 2007). The NRC (1998) requirement for total dietary arginine for pigs weighing 3 to 5 kg is 0.59 g arginine/100g feed (0.59%) and for pigs weighing 5 to 10 kg it is 0.54 g arginine/100g feed (0.54%). In the current experiment, the light weaner pigs in the control diets (no supplemental arginine) consumed on average 1.63 g arginine/day (at an average feed intake of 148g/day, which is equivalent to 1.1 g arginine/100 g of feed intake) (Table 4). Therefore the light pigs in the control treatment, without supplemental arginine were not limited in their arginine intake. In standard commercial diets it is impossible to formulate diets without arginine, as it is a naturally occurring amino acid in dietary ingredients such as grain, meat by products and milk powders.

These results differ to those found by Hernandez *et al.* (2009) who showed significant increases in daily feed intake and daily gain in the period between days 11 and 14 post-weaning. Pigs in that experiment were not deficient in arginine either and average daily arginine intakes were above NRC requirements (1.08g arginine/100g intake in control diet and 1.52 g arginine/100g in supplemented diet). There is no clear explanation as to why similar results in growth performance and feed intake were not observed in the current experiment between 12 and 20 days post-weaning. At the start of the experiment, the pigs were the same age and the two live weight classes covered the weight described by Hernandez *et al.* (2009).

The results from this experiment show that it is not necessary to provide supplementary arginine over and above NRC requirements. Naturally occurring arginine in commercial diets should be sufficient to meet arginine requirements. Ball *et al*. (2007) showed that dietary intake and *de novo* synthesis of arginine should be sufficient to meet the metabolic arginine requirement in young piglets, and that supplemental arginine to the diets of newly weaned pigs is not required. Our data from this experiment and our earlier study for Project 2B-103 (Morrison *et al*, 2009) supports this theory.

The live weight of pigs at weaning influenced the growth performance and feed intake during the weaner phase. The growth performance data during the initial 12 days post-weaning suggests that the lighter piglets at weaning may have outperformed the heavier pigs in terms of weight gain during this time, although the differences were not significant. These results were similar to those observed by the previous CRC project 2B103 (Morrison *et al.* 2009) and Collins *et al.* (2009 a,b) where the weaning weight was negatively correlated with rate of gain in the first five days post-weaning. The estimates of feed intake during this initial period post-weaning were higher in the heavy-weight class. Other investigations have reported light pigs consume more feed during the initial three days post-weaning compared to heavy weaning weight pigs (Bruininx *et al.* 2001). This may suggest that either the heavier pigs spend more time fighting and establishing social hierarchies during the initial post-weaning period than the lighter pigs or that the heavier pigs find it more difficult to adapt to the changes in environment and feed source at weaning. It is speculated that similar differences would have been observed in the current experiment if pigs were weighed three days post-weaning, and that these differences had disappeared by 10 days post-weaning.

In the current experiment, overall, the pigs that were heavier at weaning were heavier at the end of the weaner phase, and had higher average daily feed intake and daily gain compared to the light-weight weaners. Given the strong association between birth weight, weaning weight and lifetime growth performance (Morrison *et al*, 2009; Collins *et al*, 2009a,b; Dunshea *et al*, 2003) strategies to increase weaning weight are required.

5. Conclusion/Recommendations

In conclusion, this experiment suggests that supplementing additional arginine, over and above the NRC requirements does not improve growth performance and survivability of pigs weaned at 21 days of age.

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