

SUPPLEMENTATION OF LACTATING SOW DIETS WITH GLUTAMINE TO IMPROVE MILK YIELD AND GROWTH OF PIGLETS.

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Executive Summary

Glutamine is an amino acid that is abundant in plant and animal tissues. It appears to play an important role in the development of the piglet both in gestation and early growth. Glutamine is abundant in sow's milk and supports the rapid growth and maturation of the small intestine in the piglet. There have been recent recommendations to include 1 % glutamine in the diets of sows in late gestation and lactation, and the diet of newly weaned pigs. However the use of glutamine at this level is cost prohibitive. This experiment investigated the benefits that may be obtained in piglet performance during lactation when the gilt's diet is supplemented with a cheap and readily available source of glutamine.

Diets (14.5 MJ DE/kg, 0.80 g AvL/MJ DE) were offered to lactating gilts (n=50) throughout lactation (21 d). Diets were offered on a step-up basis for the first 5 days of lactation, and then gilts were fed to appetite. Gilts received one of three treatments:

1. Control diet.
2. Control diet + 1 % Glutamic acid
3. Control diet + 1.15 % Monosodium glutamate (MSG)

Litters were standardized to a common litter size and weighed on the morning after their birth (within 24 hours of birth) and again at weaning. Fostering was kept to a minimum and when required occurred within treatments. Gilt feed intake was also recorded to ensure no deleterious effects on gilt feed intake were observed.

Lactation length and gilt feed intake did not differ significantly between treatments, although those gilts that received the control diet supplemented with 1 % glutamic acid ate 9 % more feed across the lactation.

Piglet weight did not differ significantly at the start of lactation or when they were weaned. However, when treatments were compared on an average daily gain basis, piglets receiving the control diet supplemented with 1.15 % MSG grew significantly faster ($P=0.042$, 172 c.f. 188 g/d) than either the control or 1 % supplemented diet.

Glutamine would appear to be an important amino acid that has been overlooked. Recent research reviews have highlighted the role that it plays in foetal and young pig development, however, it is cost prohibitive to include glutamine at recommended levels. It is also possible that the wider range of ingredients used in Australia, especially our ability to use restricted animal material, may mean our dietary glutamine levels are higher than a typical corn-soybean diet.

This experiment investigated a cheaper source of glutamine that showed some potential to improve piglet growth performance, further validation of this potential should occur before recommendations on dietary inclusion are made.

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1. Introduction

Glutamine is an amino acid that has a long history being first obtained from beets in 1883, isolated from a protein (hemp) in 1932 and synthesized in 1933. It is an abundant amino acid found in plant and animal proteins and due to its abundance, it has not traditionally been considered a nutrient needed in livestock diets. It is found in most bulk feed ingredients utilized in this country in various concentrations (Table 1). It is found in low concentrations in cereal grains (corn, sorghum), higher concentrations in animal and protein meals (eg blood meal, fish meal, soybean meal) and very high concentrations in milk proteins (casein).

Table 1. Glutamine (Gln) concentration (% , as fed) of common feed ingredients.

Source: Li *et al.* (2011) *Amino Acids* 40: 1159-1168.

Ingredient	Gln %, as fed basis
Biscuit meal	1.4
Blood meal	4.3
Casein	11.2
Corn	1.0
Cottonseed meal	3.8
Feather meal	2.9
Fish meal	3.9
Gelatin	3.0
Meat and bone meal	2.8
Peanut meal	2.7
Poultry meal	3.5
Soybean meal	3.8
Soybean meal (dehulled)	4.1
Sorghum	0.9

There are various reasons why glutamine has not been considered important. Traditionally it has not been described as a component of proteins (Maynard *et al.*, 1979; Bondi, 1987) and is not mentioned in classical animal nutrition texts (Morrison, 1956; Cullison and Lowrey, 1987; Cheeke, 1999) and it is only with recent advancements in laboratory methods that accurate analysis has been possible. As a consequence the current (10th Edition) NRC guidelines (1998) do not recommend dietary requirements.

The abundance of glutamine within the fluids and tissues of the body suggests that it may be more important than we traditionally thought, and especially for the young piglet. A recent review by Wu *et al.* (2011) highlights some of these important roles of glutamine (Gln):

- The concentration of Gln in sow's milk is 7-fold the level in plasma.
- Gln (and glutamic acid) accounts for 20 per cent of the bound amino acids in milk and colostrum.
- Gln has been found to support rapid growth and maturation of the small intestine in the piglet.
- The concentration of Gln in the plasma of fetal pigs is 3 times that of postnatal animals.

- The concentration of Gln in amniotic and allantoic fluids is raised in critical times in the development of the fetus and the placenta, the concentration of Gln is 20 times base levels during the period 10 to 15 days post-conception. In comparison, lysine concentrations increase by a maximum of 50 per cent over gestation.

Glutamine is also known to have many other effects outside of the fetal pig (Wu *et al.*, 2011). It plays important roles in the proliferation of cells as both an energy source for rapidly dividing cells and as a precursor for purine/pyrimidine. It is a major amino acid catabolized for the synthesis of arginine within the body (the role of arginine in NOx production is thought responsible for increased milk yields of glutamine supplemented sows) and modulates gene expression regulating nutrient metabolism. When fed post-weaning it has also been seen to reduce, by 25%, the concentration of cortisol in plasma.

This information has led to various studies being conducted in the weaned pig. Johnson *et al.* (2006) fed 21 day old weaned pigs diets containing an additional 4.38% glutamine for 14 days. Whilst no effect was seen on feed intake or final weight they saw significant modifications, potentially beneficial, of immune cells in mesenteric lymph nodes. Jiang *et al.* (2010) offered sows 0 or 1 per cent glutamine throughout their lactation. They found that milk yield increased, the concentration within the milk increased two to three-fold and that the piglet grew faster pre-weaning and had a more developed gut with increased villous height and crypt depth.

Yi *et al.* (2005) compared control diets to diets supplemented with seven per cent spray dried porcine plasma (SDPP) and two per cent glutamine for 14 days post-weaning, before challenging the pigs with *E. coli* K88⁺. They found that, pre-challenge, there was no effect on average daily gain. Post the challenge they found that the feed conversion ratio was not significantly different between the SDPP, glutamine and an unchallenged control group. The challenged control, SDPP and glutamine groups all showed infection via increased rectal temperature, but only the challenged control group had reduced performance, with greatly increased villous atrophy being seen.

As a result of these studies and work of their own, Wu *et al.* (2011) have suggested that specific diets should be supplemented with one per cent glutamine. A gestation diet fed from day 90 onwards would ameliorate fetal growth retardation and pre-weaning mortality. A lactation diet containing one per cent glutamine would enhance milk production, and in creep diets it would help maintain gut health, prevent intestinal dysfunction and increase growth and survival.

This supplementation would come with significant cost. The inclusion of glutamine at one per cent would increase the cost of diets by \$200 to \$250 per tonne, but there remains the possibility that a cheaper more readily available glutamic acid donor may be able to replicate some of the results seen through glutamine use at one-tenth of the cost. This study was designed to test the hypothesis that piglet performance during lactation will be enhanced by the supplementation of the gilt's diet with a more readily available glutamic acid donor.

2. Methodology

One-hundred and fifty (n=50) gilts were offered one of three diets (14.5 MJ digestible energy (DE)/kg, 0.80 g available lysine/MJ DE) throughout lactation (21 days). The three diets (table 2) were a control treatment, a diet containing 1 per cent glutamic acid and a diet containing 1.15 per cent monosodium glutamate (MSG). The higher inclusion rate for MSG was to deliver the same concentration of glutamic acid in the diet. Salt (NaCl) was removed from the MSG diet to take account of the sodium component of MSG. Diets were offered on a step-up basis for the first 5 days of lactation, and then gilts were fed to appetite.

Table 2. Diet formulations for control treatment diets (Control), 1 % glutamic acid (L-Glu) and 1.15 % monosodium glutamate (MSG).

Ingredients (g/kg)	Control	L-Glu	MSG
Sorghum 10.0	241.0	230.5	232.0
Wheat 12.5	300.0	299.6	300.0
Chick peas 23.0	100.0	100.0	100.0
Canola meal 35.0	100.0	100.0	100.0
Soybean meal 48.0	100.0	100.0	100.0
Full-fat soybean meal 37.0	20.0	22.0	21.5
Blood meal 85.0	30.0	30.0	30.0
Meat meal 48.0	64.5	64.5	64.5
Tallow	29.0	28.0	27.0
Limestone (fine)	2.0	2.0	2.0
Salt (fine)	2.0	2.0	-
Choline chloride 60%	0.75	0.75	0.75
Betaine	2.0	2.0	2.0
DL Methionine	0.35	0.35	0.35
Lysine sulphate (51% Lysine)	3.70	3.65	3.70
L-Threonine	0.4	0.4	0.4
L-Glutamic acid	-	10.0	-
Monosodium glutamate	-	-	11.5
Optisweet	0.3	0.3	0.3
Reprotax	2.0	2.0	2.0
Vit/Min premix - breeder	2.0	2.0	2.0
<i>Analysis</i>			
DE, MJ/kg	14.5	14.5	14.5
CP, %	23.5	23.4	23.4
Lys, %	1.45	1.45	1.45
Av Lys, g/MJ DE	0.80	0.80	0.80
Salt (NaCl), %	0.41	0.41	0.22
Sodium, %	0.17	0.17	0.24

Litters were standardized to a common litter size and weighed on the morning after their birth (within 24 hours of birth) and again at weaning. Fostering was kept to a minimum and when required occurred within treatments. Gilt feed intake was also recorded to ensure no deleterious effects on gilt feed intake were observed.

A milk sample was also obtained from a subset of gilts (n=10) to observe potential changes in glutamine concentration in milk.

The litter was the experimental unit and data were subjected to an analysis of variance and means were separated by least significant differences ($P < 0.05$).

3. Results

Treatments did not differ significantly in lactation length or litter size at either the start of lactation or at weaning (table 3.). Whilst feed intake was not significantly different between treatments, those gilts that received the control diet supplemented with one per cent glutamic acid ate almost 10 per cent more feed across the lactation. This may have significant impact at a commercial level.

Table 3. Lactation length, litter size and feed intake of gilts receiving a control diet, a diet containing 1 % glutamine (L-Glu) and 1.15% monosodium glutamate (MSG).

	N.	Lactation length (d)	Litter size start	Litter size wean	Total feed intake (kg)	ADFI (kg/d)
Control	44	21.2	11.1	10.5	114.8	5.4
L-Glu	41	21.0	11.2	10.3	124.7	5.9
MSG	40	20.5	11.1	10.3	114.3	5.6
SED		0.30	0.17	0.24	7.03	0.34
P value		0.066	0.760	0.647	0.256	0.312

^{a,b,c}Means in a column with different superscripts differ significantly ($P < 0.05$); ADFI, average daily feed intake; SED, standard error of difference.

Gilts fed MSG during lactation suckled piglets with a significantly higher average daily gain than those fed a control diet or diet containing L-glutamine (table 4.). There was no significant difference in start or end weight for piglets nor was total gain significantly different, but this is likely due to the small size of this experiment relative to the parameters being investigated.

Table 4. Average individual piglet performance from gilts receiving a control diet, a diet containing 1 % glutamine and 1.15% monosodium glutamate (MSG), respectively.

	N.	Start weight (kg)	End weight (kg)	Gain (kg)	ADG (kg/d)
Control	44	1.44	5.07	3.63	172 ^a
L-glu	41	1.46	5.10	3.64	174 ^a
MSG	40	1.46	5.31	3.85	188 ^b
SED		0.050	0.160	0.139	6.8
P value		0.858	0.256	0.210	0.042

^{a,b,c}Means in a column with different superscripts differ significantly ($P < 0.05$); ADG, average daily gain; SED, standard error of difference.

4. Outcomes

The supplementation of gilt lactation diets with 1.15 per cent MSG did not result in any deleterious effects on the sow, with her feed intake being maintained. Within the piglets suckling these gilts the average daily gain of pigs suckling mothers who were fed MSG increased significantly.

The experiment investigated a cheaper source of glutamic acid that showed some potential to improve piglet growth performance. Further investigation is required to confirm and exploit the potential benefits of adding supplementary glutamine to gilt and sow diets during lactation and potentially to piglet diets post-weaning.

5. Application of Research

The supplementation of gilt diets with MSG appears to be a possible method to increase piglet performance prior to weaning, however, further investigation is required to exploit these potential benefits. The review of literature conducted during this project highlighted the importance of glutamine in the growth and development of the pig. The recommendations of Wu *et al.* (2011) to include one per cent glutamine into the late

gestation, lactation and early diets for weaned piglets do not appear economically feasible. Given these recommendations are based on corn-soybean diets, the investigation of glutamine levels in Australian diets, where we utilize high source products such as meat and bone meal, may be an initial step taken to check the value of these recommendations to the Australian pig industry.

6. Conclusion

This project has delivered:

- Increased piglet growth rates through the supplementation of the lactating gilts diet with MSG.
- A treatment that increased piglet growth rate that did not have a deleterious effect on lactation feed intake.

7. Limitations/Risks

The use of MSG does increase the sodium content of the diet, even with the removal of salt (sodium chloride) from the formulation, so care should be taken where stock water is highly sodic. Our multi-ingredient diets are possibly different to the corn-soybean diet in glutamine content. From an industry perspective it would appear imperative to sample diets for current glutamine content before embarking on a program of supplementation.

8. Recommendations

Glutamine would appear to be an important amino acid that has been overlooked. Recent research reviews have highlighted the role that it plays in foetal and young pig development, however, it is cost prohibitive to include glutamine at recommended levels.

This experiment investigated a cheaper source of glutamic acid that showed some potential to improve piglet growth performance. Further validation of this potential should occur before recommendations on dietary inclusion are made. The direct feeding of diets containing MSG to weaned piglets may also be a possible method to enhance gut health and reduce the post-weaning growth check.

Similarly it is also possible that the wider range of ingredients used in Australia, especially our ability to use restricted animal material, may mean our dietary glutamine levels are higher than a typical corn-soybean diet. Given differences in diet formulations in Australia, it may be wise to conduct a survey of finished feeds to assess their glutamine concentrations.

9. References

- Bondi, A.A. (1987) *Animal Nutrition*. John Wiley and Sons, New York, NY.
- Cheeke, P.R. (1999) *Applied Animal Nutrition: Feeds and Feeding*. Prentice Hall, Upper Saddle River, NJ.
- Cullison, A.E. and Lowrey, R.S. (1987) *Feeds and Feeding*. 4th Edition. Prentice Hall, Englewood Cliffs, NJ.
- Johnson, I.R., Ball, R.O., Baracos, V.E. and Field, C.J. (2006) Glutamine supplementation influences immune development in the newly weaned piglet. *Development and Comparative Immunology* 30:1191-1202.
- Li, X., Rezaei, R., Li, P. and Wu, G. (2011) Composition of amino acids in feed ingredients for animal diets. *Amino Acids* 40: 1159-1168.
- Maynard, L.A., Loosli, J.K., Hintz, H.F. and Warner, R.G. (1979) *Animal Nutrition*. 7th Edition. McGraw-Hill Book Co., New York, NY.
- Morrison, F.B. (1956) *Feeds and Feeding*. 22nd Edition. The Morrison Publishing Co., Ithaca, NY.
- NRC (1998) *Nutrient Requirements of Swine*. 10th Edition. National Academy Press, Washington, DC.
- Wu, G., Bazer, F.W., Johnson, G.A., Knabe, D.A., Burghardt, R.C., Spencer, T.E., Li, X.L. and Wang, J.J. (2011) *Journal of Animal Science* 89:2017-2030.
- Yi, G.F., Carroll, J.A., Allee, G.L., Gaines, A.M., Kendall, D.C., Usry, J.L., Toride, Y. and Izuru, S. (2005) Effect of glutamine and spray-dried plasma on growth performance, small intestine morphology, and immune responses of Escherichia coli K88+-challenged weaned pigs. *Journal of Animal Science*. 83: 634-643