Reducing sucker mortality through a novel in feed acid during late gestation and lactation

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Report prepared for the
Co-operative Research Centre for High Integrity Australian Pork

By

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Executive Summary

High pre-weaning mortality is still a major concern for pork producers, with *Escherichia* coli causing significant ill thrift and mortality at this age. The development of new and innovative feed additives to reduce the microbial transfer of *E.coli* and other microbial pathogens such as *Salmonella* spp. from sows to piglets has potential to be used as a component in an integrated health program. Mannose-containing oligosaccharides (MOS) have been used as an aid against pathogenic bacteria in pig and poultry diets with increasing popularity. ß1-4 mannobiose (MNB) is an indigestible disaccharide derived from copra meal by enzymatic digestion. Initial investigations have suggested MNB may act to increase the gram negative killing activity of macrophages and may also up-regulate the expression of genes involved in host-defense. In addition, there is evidence that MNB can inhibit gram negative bacterial adhesin to the intestinal cell by up to 80%. The aim of this project was to facilitate the development and assessment of a new in feed additive containing high levels of ß1-4 mannobiose in combination with a mix of synergistic organic acids. The new product will be assessed for its ability to modify key microbial populations in the sow as well as its ability to reduce pre-weaning mortality.

A total of 688 primiparous sows (Large White x Landrace) were allocated to one of four dietary treatments at 14 weeks of gestation: A: Control (no acidifiers); B: 6 kg/t Fysal™ (blend of short chain fatty acids); C: 4 kg/t Fysal Fit4™ (blend of short and medium chain fatty acids plus low dose ß1-4 mannobiose); D: 4 kg/t Fysal Fit4 Plus (blend of short and medium chain fatty acids plus a high dose ß1-4 mannobiose). Diets were fed from 14 weeks of gestation and throughout lactation. Measures of litter growth performance and mortality were obtained along with measurements of sow feed intake during lactation. Faecal samples were obtained from a subset of sows at 9 weeks of gestation, 15 weeks of gestation and again during the second week of lactation for microbiological enumeration for *Clostridium perfringens,* coliforms, *E.coli, Lactobacillus* species and *Enterobacteriaceae*.

Pre-weaning mortality in the control sows was 16.9% compared to 16.2 %, 18.7 % and 15.7% for the Fysal™, Fysal Fit 4™ and Fysal Fit4 Plus treatment groups (x^2= 5.50, P=0.14). Piglet growth rates and litter weaning weights were similar amongst treatments, while sow feed intake was modestly improved when sows were offered the Fysal Fit4 Plus diets. Microbial analysis of the faecal samples collected at mid-lactation showed distinct reductions in *E.coli* counts when sows had been offered one of the acidified gestation and lactation diets, with the counts numerically lowest in the Fysal Fit4 Plus treatment group.

The outcomes from this study confirm that the inclusion of Fysal Fit4 plus in late gestation and lactation diets can reduce faecal microbial populations in a similar manner to Fysal™ or Fysal Fit4™. Numerical improvements in pre-weaning mortality were observed, however further investigation with a larger number of mixed parity sows would be warranted to confirm and quantify the benefits to producers above currently available products.

Table of Contents

1. Introduction

Enterotoxigenic *Escherichia* coli (*E.coli*) are severe pathogens in Australian pig herds, causing significant ill thrift and mortality in both sucker and weaner pigs. Worldwide the O157 *E. coli* are extreme pathogens and experienced practitioners rely on a mix of hygiene, acids, environment, and pig flows to minimize the incidence of disease. The development of new and innovative feed additives to reduce the microbial transfer of *E.coli* and other microbial pathogens such as *Salmonella* spp. from sows to piglets has strong potential to be used as a component in an integrated health program.

The application of organic acids and their salts to pig diets in an effort to reduce the targeted microbial population and the incidence of disease has been studied extensively. Their inclusion in feed reduces the feed’s buffering capacity and pH, while their antimicrobial effect helps prevent the growth of bacteria (especially gram negative bacterial species such as *Salmonella* spp. and *E.coli*). Their mode of action can be explained by the decrease in environmental pH and by the ability of these acids to pass the cell membrane where they dissociate and disturb the metabolism of the microbe.

In addition to acidifiers, a number of oligosaccharide supplements are now commercially available for use in feed to improve the immune response against pathogens. Mannose-containing oligosaccharides (MOS) have been used as an aid against pathogenic bacteria in both pig (LeMieuz et al., 2003)and poultry diets (Fernandez et al., 2002) with their popularity increasing as producers aim to reduce the impact of disease. A number of different oligosaccharides have emerged that have variable effects on general health and immune responses to pathogens in both humans and animals. ß1-4 mannobiose (MNB) is one such compound, an indigestible disaccharide derived from copra meal by enzymatic digestion. Mannobiose appears to have several modes of action all working to reduce the impact of pathogenic bacteria on the host animal. Firstly, MNB is a potential inhibitor of FimH adhesin in gram negative bacteria, blocking adhesin of the bacteria to the intestinal cell. In addition, MNB may also improve the gram negative-killing activity of macrophages, and may act as a potent immune-modulator via its ability to up-regulate the expression of genes involved in host-defense (Ibuki *et al*. 2010).

The mechanism by which MNB blocks the adhesin of gram negative bacteria to the intestinal cells appears to involve the mannose-dependent or mannose-sensitive adhesin through type 1 fimbrae. Other molecules that interfere with the surface structure of gram negative bacteria and/or the enterocyte may also affect the binding process. In vitro models to study the adhesin inhibition of gram negative *salmonella* spp. to a monolayer of a defined pig jejunal enterocyte cell line, denoted as IPEC-J2 cells are now available (Scheirack *et al*. 2006). Using this cell line, MNB has been measured to inhibit adhesin of *Salmonella enteritidis* and *Salmonella Typhimurium* by 50-80% (Nutreco R&D Trial Summary ASG-2010).

This project will facilitate the development of an in feed additive containing high levels of ß1-4 mannobiose in combination with a mix of synergistic organic acids. The new product will be specifically targeted to reduce the adhesin of virulent *E.coli* in the intestine of sucker pigs and will be assessed for its ability to reduce pre-weaning mortality and ill thrift compared to other organic acid blends currently available in Australia. In addition, faecal samples obtained directly from the sow throughout the feeding period will provide an indication of how the in-feed products are manipulating microbial populations.

2. Methodology

Animals and treatments

A total of 688 primiparous sows (Large White x Landrace) were selected during pregnancy and housed in pens of two sows/pen (1.6 m^2/sow). Sows were selected over 13 time replicates (weeks), with approximately 40 – 60 sows selected each week depending on production flows. At 14 weeks of gestation sows were randomly allocated within replicate to one of four dietary treatments:

A. Control gestation and lactation diets (no acidifiers)
B. Gestation and lactation diets containing 6 kg/t Fysal™(blend of short chain fatty acids, Nutreco, The Netherlands)
C. Gestation and lactation diets containing 4 kg/t Fysal Fit4™ (blend of short and medium chain fatty acids plus low dose ß1-4 mannobiose, Nutreco, The Netherlands)
D. Gestation and lactation diets containing 4 kg/t Fysal Fit4 Plus (blend of short and medium chain fatty acids plus a high dose ß1-4 mannobiose)

The ingredient profile and nutritional composition of the gestation and lactation diets are displayed in Tables 1 and 2. The gestation diet was offered from 14 weeks of gestation through to farrowing, with the lactation diet fed from farrowing through to weaning. During gestation, sows were floor fed a total of 2.4 kg/d until farrowing, after which time the lactation diet was offered using a step up program to *ad libitum* intakes by day 5. All animal procedures outlined in this investigation were approved by the Rivalea Animal Ethics committee.

Table 1 - Ingredient composition and nutrient profile of each of the gestation diets, % of diet (as fed basis).

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 plus
Rolled Wheat	40.0	40.0	40.0	40.0
Barley	24.4	24.4	24.4	24.4
Mill mix	14.9	14.9	14.9	14.9
Hull mixture	2.5	2.5	2.5	2.5
Canola meal	5.0	5.0	5.0	5.0
Meat meal	2.5	2.5	2.5	2.5
Fish meal	1.0	1.0	1.0	1.0
Semi refined fish oil	0.3	0.3	0.3	0.3
Tallow	5.0	5.0	5.0	5.0
Salt	0.35	0.35	0.35	0.35
Limestone	0.67	0.67	0.67	0.67
Dicalcium phosphate	0.33	0.33	0.33	0.33
Lysine HCL	0.40	0.40	0.40	0.40
DL-Methionine	0.09	0.09	0.09	0.09
Magnesium sulphate	0.60	0.60	0.60	0.60
Threonine	0.16	0.16	0.16	0.16
Endox	0.02	0.02	0.02	0.02
Phytase	0.07	0.07	0.07	0.07
Arbocel	1.33	1.33	1.33	1.33
Levucell SB	0.02	0.02	0.02	0.02
Vitamin and mineral premix	0.84	0.84	0.84	0.84
Fysal™		0.6		
Fysal Fit4™			0.4	
Fysal Fit 4 Plus				0.4
*Estimated nutrient composition, %**				
DE, MJ/kg	13.8	13.8	13.8	13.8

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 plus
Crude protein	13.6	13.6	13.6	13.6
Crude fat	6.9	6.9	6.9	6.9
Crude fibre	6.0	6.0	6.0	6.0
Available lysine: DE ratio g/MJ DE	0.54	0.54	0.54	0.54

Table 2 -Ingredient composition and nutrient profile of each of the lactation diets, % of diet (as fed basis).

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 plus
Rolled Wheat	57.2	57.2	57.2	57.2
Barley	11.5	11.5	11.5	11.5
Mill mix	5.6	5.6	5.6	5.6
Soyabean meal	2.0	2.0	2.0	2.0
Canola meal	7.5	7.5	7.5	7.5
Meat meal	55.1	55.1	55.1	55.1
Blood meal	0.7	0.7	0.7	0.7
Semi refined fish oil	0.4	0.4	0.4	0.4
Tallow	6.6	6.6	6.6	6.6
Liquid betaine	0.4	0.4	0.4	0.4
Salt	0.35	0.35	0.35	0.35
Limestone	0.83	0.83	0.83	0.83
Lysine HCL	0.38	0.38	0.38	0.38
DL-Methionine	0.11	0.11	0.11	0.11
Magnesium sulphate	0.60	0.60	0.60	0.60
Threonine	0.17	0.17	0.17	0.17
Ronozyme	0.03	0.03	0.03	0.03
Endox	0.02	0.02	0.02	0.02
Phytase	0.07	0.07	0.07	0.07
Vitamin and mineral premix	1.22	1.22	1.22	1.22
Fysal™		0.6		
Fysal Fit4™			0.4	
Fysal Fit 4 Plus				0.4
*Estimated nutrient composition, %**				
DE, MJ/kg	14.9	14.9	14.9	14.9
Crude protein	16.2	16.2	16.2	16.2
Crude fat	8.5	8.5	8.5	8.5
Crude fibre	3.6	3.6	3.6	3.6
Available lysine: DE ratio g/MJ DE	0.57	0.57	0.57	0.57

Husbandry and measurements

Sows were housed in the gestation facility until approximately 110 days of gestation, at which time they were moved into conventional farrowing crates in an insulated shed. Not all animals offered the treatment diets during gestation remained on the study throughout lactation due to pregnancy fall out and production flow constraints. A total of 583 sows were moved into the farrowing house facility for measurement of the treatment effects on litter survival and growth performance.

Sows farrowed naturally at term, with litters processed within 24 hrs of birth. Litter size was standardized where necessary to the maximum number of functional teats using a minimal cross fostering approach with piglets born on the same day.Feed intake was measured on a per sow basis from farrowing through to weaning using calibrated scoops. Litter weight was measured after fostering and again just before weaning to determine growth performance of the litter. All pre-weaning piglet and sow mortalities and removals were recorded including the reason and date. In addition, a scour score was obtained on all litters in replicates 3 onwards as an indication of litter health. A simple scoring system was used: 0 – no scouring present; 1 – low level scouring (pasty); 2- medium level scouring (sloppy); 3-high level scouring (watery). Scours scores were obtained between 7 and 10 days of lactation.

Faecal samples were obtained from a small subset of sows at 9 weeks of gestation, 15 weeks of gestation and again during the second week of lactation (approximately 15 sows per dietary treatment, the same sows sampled at all three time points). Samples were collected directly from the sow into sterile bags, frozen and subsequently transported to ACE laboratories, Bendigo, Victoria for microbiological enumeration for *Clostridium perfringens,* coliforms, *E.coli, Lactobacillus* species and Enterobacteriaceae.

Statistical analyses

The influence of dietary treatment on the sow's lactation performance was analysed using REML with the main effect of treatment and the random effect of replicate (week). Covariates were included in the model where there were significant effects and are indicated in the text and footnotes of the tables. Chi-squared analysis was undertaken to determine differences in mortalities and removals across the four treatments groups as well as for the analysis of some of the subsequent reproduction data. All statistical analyses were undertaken using GenStat 16th Edition (Payne *et al.* 2005).

3. Outcomes

Sow characteristics

Of the 688 sows allocated to one of the four dietary treatments, a total of 583 were followed through lactation for measurement of litter performance and survival. The main reason for a sow being omitted before lactation was due to her being identified as not in pig or due to other production flow constraints. The higher than usual percentage of sows not in pig was due to the study running over the summer infertility period, with sows mated from late December through to late April. The study utilized primarily nucleus gilts, with the distribution of genetic lines similar across treatments (Table 3). A small number of commercial gilts were also included, with the distribution of these very similar across dietary treatments.

The K88 genetic status of the nucleus gilts and boars were for the most part known, with the mating distribution across treatments displayed in Table 4. Single sire matings were designed to produce predominantly resistant offspring, with the resistant combinations dominating as shown in Table 4. There were a similar number of K88 resistant and susceptible genetic combinations across the four dietary treatment groups (x^2= 3.73, P=0.30). The influence of dietary treatment on sow mortality and removal during lactation is displayed in Table 5. There was no effect of treatment on the number of deaths and removals (excluding removals for 'other' reasons, x^2= 5.36, P=0.15), although the control treatment was the only one to have any sows die during lactation. Removal of the sow and her litter from the study for 'other' reasons was mainly due to issues with boards between two farrowing crates coming down prior to weaning and the litters being mixed.

Table 3 - Distribution of nucleus sows across treatments

	Number sows entering farrowing house			
	Line 1	Line 2	Line 5	Commercial
Control	59	66	14	9
Fysal	56	61	21	7
Fysal Fit4	69	54	13	9
Fysal Fit4 Plus	58	61	18	8

Table 4 - Distribution of K88 status across each of the dietary treatment groups

	Sow K88 Status (sow/boar)															
	Resistant combinations							Susceptible combinations			Other					
Treatment	SR	RC	CR	RR	RU	UR	Total Resistant combinations	SC	CC	Total susceptible / carrier	CU	SU	UU	UC	Total other	TOTAL
Control	15	18	49	31	1	1	115	2	19	21	1	0	9	2	12	148
Fysal™	12	14	44	32	0	2	104	6	22	28	2	1	8	2	13	145
Fysal Fit4™	15	18	33	29	0	4	99	9	23	32	1	1	10	2	14	145
Fysal Fit4 Plus	14	23	45	27	0	2	111	5	20	25	0	0	8	1	9	145

S – sensitive, C – carrier, R – resistant, U- unclassified (pooled semen or sow not tested)

Table 5 - Influence of dietary treatment on sow deaths and removals (day of lactation in parenthesis)

	Sudden death	Condition	Used as foster sow	Unwell	Savaged litter	Other (management)	Total sow removals /deaths
Control	4 (day 1,3,18,25)	1 (day 19)				8	13
Fysal™			1 (day 1)	3 (day 0, 1, 2)		10	14
Fysal Fit4™						12	12
Fysal Fit4 Plus					1 (day 0)	4	5

Piglet survival and litter performance

The influence of dietary treatment on sow and litter performance is displayed in Table 6. There were no significant effects of dietary treatment on litter characteristics of born alive, still born or mummified piglets. Post foster litter size and weight were similar across treatments as were the weaning characteristics. Total litter weight at weaning was greatest when sows were offered either Fysal™ or Fysal Fit4 Plus during late gestation and lactation, although not significantly greater than the other two treatments. Scour scores measured during the second week of lactation were generally low and similar across the dietary treatments. Of interest, sow average daily feed intake was enhanced during lactation when sows were offered the Fysal Fit4 Plus treatment compared to the control diet.

The influence of dietary treatment on piglet survival before and after fostering is displayed in Table 7. Deaths are broken down into main production causes – overlays, non-viable piglet, unthrifty, savaged, hemolytic *E.coli* and scours. All recorded causes were observational only and were not confirmed by veterinary autopsy. The total number of deaths from birth to fostering was similar between the treatment groups (x2= 4.18, P=0.24). Similarly, diet did not significantly influence mortality during the post fostering to weaning period (x2= 5.50, P=0.14), although numerically pre-weaning mortality was lowest when sows were offered the Fysal Fit4 Plus treatment.

Table 6 - Influence of dietary treatment on sow and litter performance

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus	SED	P-Value
Gestation length	116.1	116.5	116.4	116.3	0.168-0.170	0.071
Born alive	10.5	10.2	10.7	10.6	0.033	0.40
Still born	0.61	0.54	0.45	0.65	0.105-0.106	0.27
Mummies	0.14	0.16	0.11	0.11	0.052-0.053	0.75
Litter size post foster	10.7	10.5	10.7	10.7	0.18-0.19	0.54
Average piglet weight post foster (kg)	1.37	1.36	1.32	1.37	0.026-0.027	0.20
Litter size weaning^	8.9	8.8	8.8	9.1	0.23-0.24	0.74
Average piglet weight weaning (kg)*	5.20	5.37	5.17	5.26	0.133-0.137	0.49
Total litter weaning weight**	47.3	48.2	46.1	48.4	1.61-1.65	0.48
Average weaning age (days)	22.4	22.1	21.9	22.3	0.26-0.27	0.28
Average daily piglet weight gain (kg/d)*	0.170	0.180	0.173	0.173	0.0051-0.0052	0.29
Scour score week 2 of lactation	0.81	0.86	0.94	0.84	0.090-0.091	0.54
Average daily sow feed intake (kg/d)	6.41	6.42	6.48	6.50	0.037-0.038	0.031

^Litter size post foster included as a covariate *Average weight post foster included as a covariate ** Litter weight post foster included as covariate

Table 7 - Influence of dietary acidifier on piglet survival through to weaning

	Pre-foster mortality (birth to processing)					Post-foster mortality (processing to weaning)					
	Overlay	Non-viable	Unthrifty	Savaged	Total (% of BA)	Overlay	Unthrifty	*E.coli*	Scours	Other	Total (% piglets post fostering)
Control	30	2	4	9	45/1549 (2.9%)	92	84	57	18	16	267/1579 (16.9%)
Fysal™	45	5	1	1	52/1476 (3.5%)	68	95	58	17	7	245/1514 (16.2%)
Fysal Fit4™	28	8			36/1564 (2.3%)	105	80	54	32	22	293/1560 (18.7%)
Fysal Fit4 Plus	26	7		9	42/1531 (2.7 %)	71	82	60	21	8	242/1535 (15.7%)

Microbial enumeration

Faecal samples collected at 9 and 15 weeks of gestation as well as week 2 of lactation were analysed for microbial counts of *E.coli*, *Lactobacillus* spp., coliform and total enterobacteriaceae. *E.coli* counts at 9 weeks of gestation were similar across sows irrespective of K88 status (Table 8). *E.coli*, coliform and total enterobacteriaceae counts were similar across dietary treatments at week 9 of lactation (Table 9), however there were some differences in *Lactobacillus* and *Clostridium* perfringens counts presumably due to natural variation between sows (feeding of the dietary treatments had not yet commenced). As such, microbial counts at week 9 of gestation were used as covariates for the subsequent two sampling points.

Samples collected prior to farrowing (15 weeks of gestation) contained similar *E.coli*, *Lactobacillus* spp., coliform and total enterobacteriaceae counts across the four dietary treatments. There was a trend for reduced *Clostridium* at 15 weeks of gestation when sows had been offered the Fysal Fit4™ treatment group (P=0.079). Analysis of faecal samples during mid-lactation showed distinct reductions in *E.coli* counts when sows had been offered the Fysal Fit4 Plus gestation and lactation diets, with the difference most predominant compared to the sows fed the control diet. In addition, pigs offered the Fysal Fit4 Plus diets also had significantly lower counts of coliforms and total enterobacteriaceae compared to the sows fed the control diets. Total *Lactobacillus* counts did not differ between treatment groups at this sampling point, while *Clostridium perfringens* was reduced in faecal samples obtained from sows offered the Fysal™ diets.

Table 8 - Influence sow K88 status on microbial counts (log10) at 9 weeks of gestation

	Resistant (n=26)	Carrier (n=25)	Susceptible (n=8)	Unclassified (n=2)	SED	P-value
E.coli	3.006	3.049	3.530	2.632	0.27-0.76	0.71
Lactobacillus spp.	8.017	8.016	7.963	7.987	0.11-0.29	0.92
Coliform	3.480	3.493	3.940	3.279	0.24-0.66	0.68
Enterobacteriaceae	3.686	3.523	4.040	3.525	0.22-0.61	0.54

Table 9 - Influence of dietary treatment on log microbial counts in sow faeces at 9 weeks gestation, 15 weeks gestation and week 2 of lactation

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus	SED	P-Value
E.coli						
Week 9 gestation	3.256	2.755	3.084	3.248	0.34-0.36	0.46
Week 15 gestation^	3.349	2.805	3.114	2.855	0.47-0.49	0.65
Week 2 lactation^	4.484	3.095	2.960	2.592	0.51-0.53	0.004
Lactobacillus spp.						
Week 9 gestation	7.775	8.148	8.180	8.010	0.12-0.13	0.009
Week 15 gestation^	7.503	7.859	7.882	7.806	0.24-0.28	0.51
Week 2 lactation^	7.041	7.468	7.180	7.483	0.24-0.27	0.23
Coliform						
Week 9 gestation	3.510	3.454	3.652	3.538	0.31-0.32	0.93

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus	SED	P-Value
Week 15 gestation^	3.696	3.413	3.377	3.686	0.37-0.39	0.77
Week 2 lactation^	4.675	3.454	3.440	2.878	0.45-0.47	0.003
Enterobacteriaceae						
Week 9 gestation	3.822	3.540	3.707	3.616	0.287-0.301	0.79
Week 15 gestation^	3.777	3.412	3.782	3.983	0.39-0.41	0.56
Week 2 lactation^	4.791	3.597	3.641	2.988	0.408-0.425	<0.001
Clostridium perfringens						
Week 9 gestation	4.959	4.803	4.380	4.774	0.203-0.213	0.048
Week 15 gestation^	4.389	4.023	3.663	4.307	0.28-0.30	0.079
Week 2 lactation^	5.359	4.040	4.628	4.732	0.33-0.36	0.003

^ Bacterial count at week 9 gestation included as a covariate in the analysis

The influence of dietary treatment on the ratio between key microbial populations is displayed in Figures 1 through to 4. The ratio of *E.coli* and *Clostridium* to *Lactobacillus* spp. (either together – figure 1, or individually – figures 2 and 3) was significantly reduced in all three acidifier treatment groups at week 2 of lactation. The ratio of *E.coli* to the total enterobacteriaceae population is displayed in Figure 4. The results between treatment groups are variable, however there was a slight trend for a reduced ratio at 15 weeks of gestation (P=0.13) when sows were offered the Fysal Fit4 Plus diets.

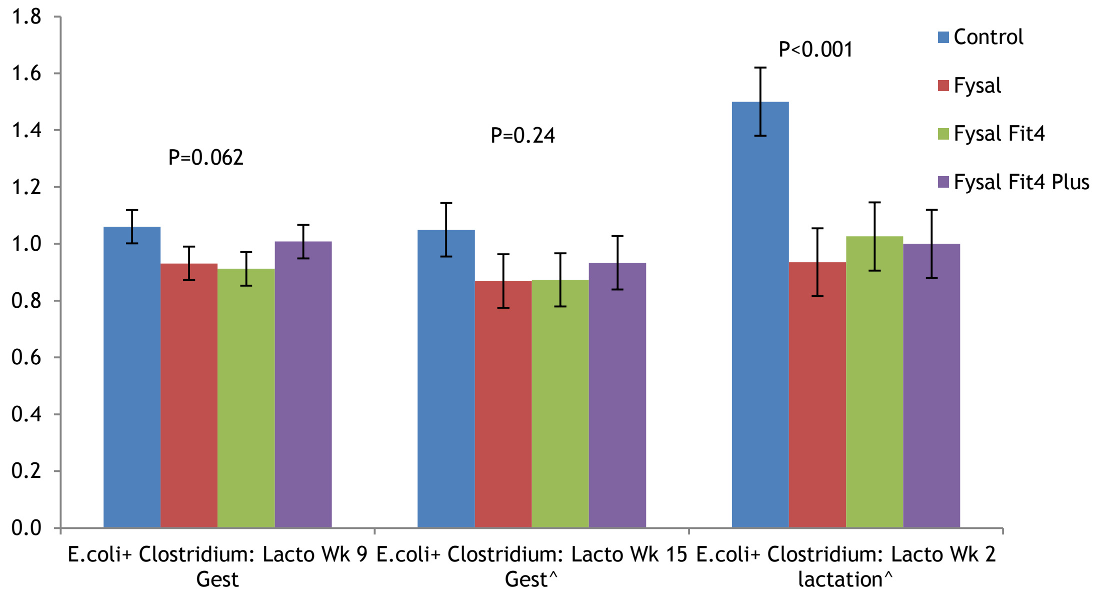

Figure 1 - Influence of dietary treatment on the ratio of *E.coli* and *Clostridium* to *Lactobacillus* spp. at 9 and 15 weeks of gestation and again at week 2 of lactation. ^Ratio at week 9 of gestation included as a covariate

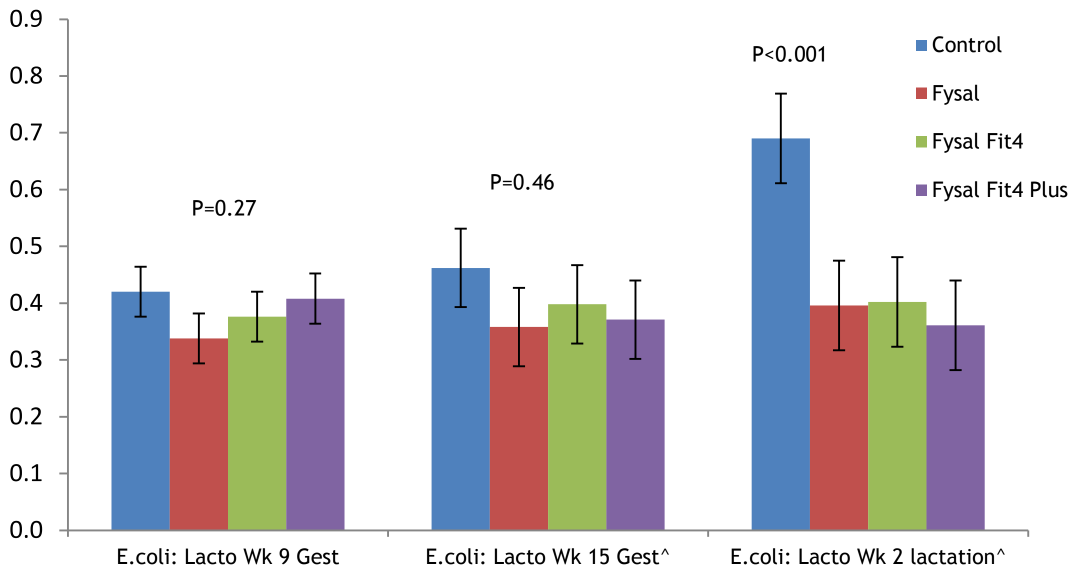

Figure 2 - Influence of dietary treatment on the ratio of *E.coli* to *Lactobacillus* spp. at 9 and 15 weeks of gestation and again at week 2 of lactation. ^Ratio at week 9 of gestation included as a covariate

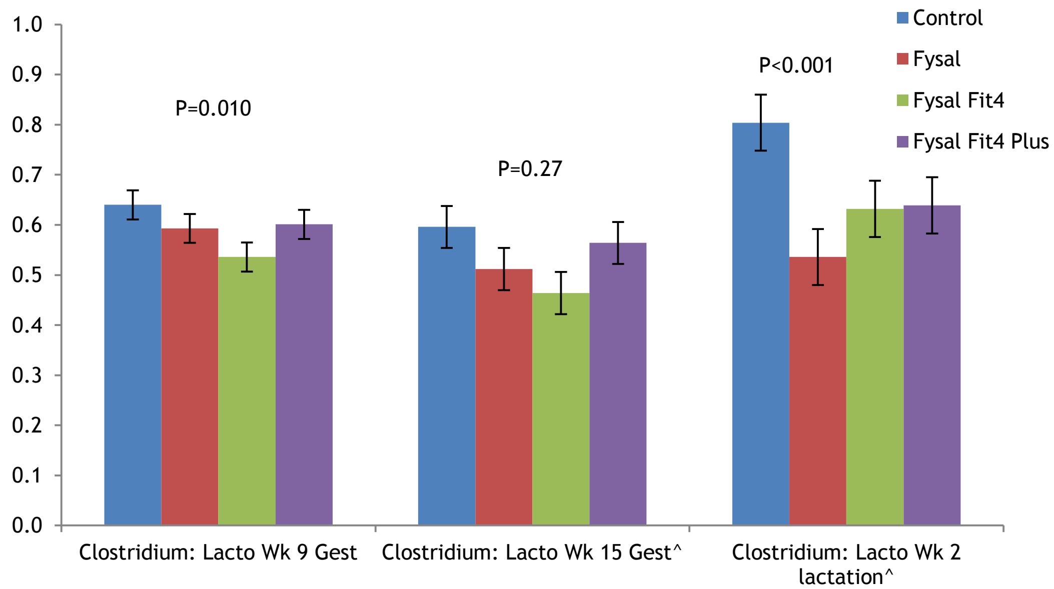

Figure 3 - Influence of dietary treatment on the ratio of *Clostridium* to *Lactobacillus* spp. at 9 and 15 weeks of gestation and again at week 2 of lactation. ^Ratio at week 9 of gestation included as a covariate

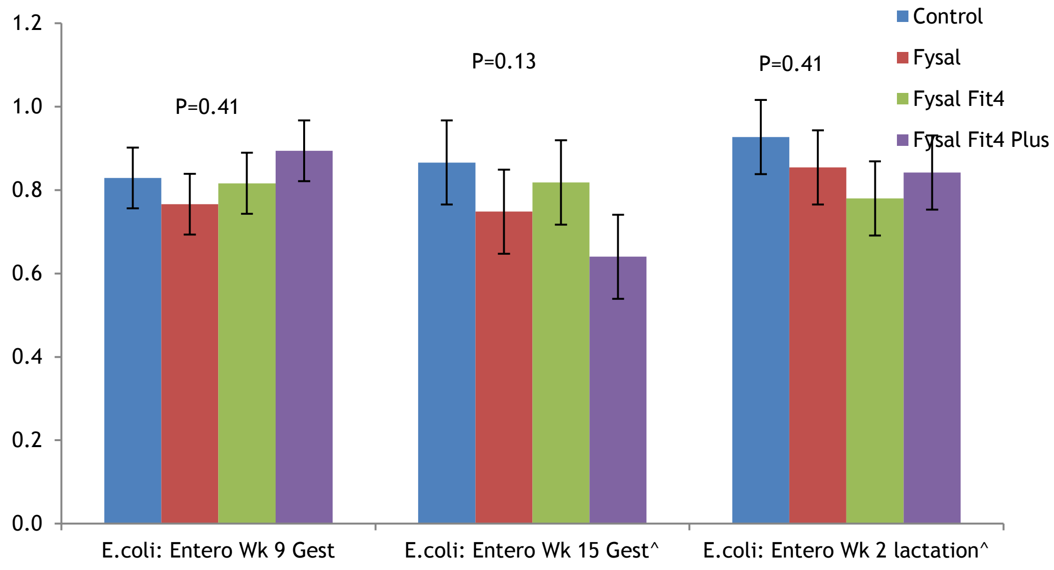

Figure 4 - Influence of dietary treatment on the ratio of *E.coli* to *enterobacteriaceae* at 9 and 15 weeks of gestation and again at week 2 of lactation. ^Ratio at week 9 of gestation included as a covariate

Subsequent reproductive performance

Subsequent reproductive performance was obtained from the herd recording system. All sows returned to standard production diets after weaning and were managed as commercial sows. A breakdown of sow removals after weaning is displayed in Table 10 along with the reason or cause. Removals were lowest in the Fysal™ treatment group, predominately due to a reduced number of sows being removed for feet/leg issues. Differences between treatments were however not significant (x2=5.48, P=0.14).

Table 10 - Influence of dietary treatment on sow removals between weaning and re-mating.

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus
Culled				
Condition	2	1	3	1
Feet and legs	8	2	8	11
Management	1			1
Stale	2	3	4	4
Sudden death	2			
TOTAL	15/135 (11.1 %)	6/131 (4.6 %)	15/133 (11.3 %)	17/140 (12.1 %)

The impact of dietary acidifier on subsequent reproductive performance is displayed in Table 11. The average number of days between weaning and re-mating was greater in the control and Fysal Fit4 Plus sows compared to those fed the standard Fysal Fit 4 diets. This was due to a greater number of sows in these treatment groups with a weaning to re-mating interval greater than 30 days (Control – 6 sows, Fysal™ – 4 sows, Fysal Fit4™ – 1 sow and Fysal Fit4 Plus – 6 sows, x2= 3.98, P=0.26). Weaning to re-mating intervals were similar when these outliers where removed from the data set (P=0.34). An overview of outcomes from the first mating after weaning is displayed in Table 12. Farrowing rate was statistically similar between dietary treatments (x2= 2.46, P=0.48), albeit numerically lower in the control group to a magnitude that would be of commercial concern. There was no difference in subsequent litter size (born alive – 11.8, 11.6, 11.8 and 11.8 for the control, Fysal™, Fysal Fit4™ and Fysal Fit4 Plus treatment groups respectively, P=0.92).

Table 11 - Influence of dietary treatment on subsequent reproductive outcomes

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus	SED	P-Value
Days weaning to re-mating	9.8	8.5	6.0	9.7	1.39-1.44	0.020
Days weaning to re-mating (excluding sows with WRI > 30 days)	7.3	7.4	6.2	7.0	0.78-0.81	0.34

Table 12 - Influence of dietary treatment on mating outcome (first mating after weaning)

	Control	Fysal™	Fysal Fit4™	Fysal Fit4 Plus
Farrowed	99	107	105	108
Returned	10	6	5	7
Pregnancy test negative	1	4	3	3
Not in pig	6	4	1	5
Sudden death	2	2	1	
Destroyed	2	2	3	
Farrowing rate (% mated)	99/120 (82.5%)	107/125 (85.6%)	105/118 (89.0%)	108/123 (87.8%)

4. Application of Research

The outcomes from this investigation suggest that the incorporation of MNB in feed during late gestation and lactation has the potential to modulate microbial populations in the sow. The microbial enumeration analysis clearly showed a reduction in total *enterobacteriaceae*, coliform and *E.coli* populations during the second week of lactation when sows had been offered feed containing Fysal Fit4 Plus compared to the control group. Faecal microbial counts of total *enterobacteriaceae*, coliform, *E.coli* and *Clostridium perfringens* were also reduced with Fysal™ and Fysal Fit4™ consumption compared to the control treatment group, again supporting the use of acidifiers for control of disease causing microbial populations in the farrowing shed.

The microbial counts obtained in this investigation were within expectation for a commercial production herd (van der Heijden *pers. comm.*). The *Clostridium* counts obtained in this investigation would be regarded as low, while the *Lactobaccilus spp.*, coliform and *Enterobacteriaceae* were also within normal expectations (van der Heijden *pers. comm.*).

The results for Fysal Fit4 Plus suggest that the presence of a higher concentration of MNB in the intestine was successful in reducing the total *E.coli* microbial population over the feeding period. There appears to be several possible modes of action by which MNB can reduce targeted microbial populations, although the exact modes of action are yet to be elucidated. Firstly, MNB may act to increase the gram negative killing activity of macrophages and may also act as a potent immunomodulator by up-regulating the expression of genes involved in host-defense. Studies in poultry have shown significantly higher levels of fecal IgA following oral administration of MNB three times a week, as well as the up-regulation of genes involved in antigen recognition and host defense (Ibuki *et al.* 2010). Secondly, there is evidence that β1-4 mannobiose may block the adhesin of gram negative bacteria to the intestinal wall, with previous studies reporting up to 80% intestinal adhesin inhibition of *Salmonella spp*. (Nutreco R&D Trial Summary ASG-2010). Together, these mechanisms appear to have reduced total gram negative bacteria in the faecal samples over time from the sows offered the Fysal Fit4 Plus diet.

The changes in microbial populations correlated with numerical reductions in pre-weaning mortality, with the lowest piglet mortality levels on the sows offered the Fysal Fit4 Plus diets. Further investigation with much larger sow numbers would be needed to confirm this effect in a multi-parity herd. The higher feed intakes from sows offered the Fysal Fit4 Plus treatment group also provides support for improved intestinal health of the sow.

The outcomes from this investigation would need to be confirmed and quantified on a mixed parity farm to assess the overall benefits to cost of production. The benefits of Fysal Fit4 plus should be considered in comparison to the currently available products – Fysal™ and Fysal Fit4™. In this study, the use of Fysal in the late gestation and lactation diets also reduced key microbial populations in the faeces and resulted in the lowest sow removal rate between weaning and re-mating. Given this, the added benefits from Fysal Fit4 Plus appear marginal and the continued use of Fysal would be advocated at this time.

5. Conclusion

The outcomes from this investigation suggest that the dietary inclusion of Fysal Fit4 Plus during late gestation and throughout lactation may reduce the population of gram negative bacteria. Pre-weaning mortality was reduced from 16.9 % in the control sows to 15.7 % in the sows offered Fysal Fit4 Plus, while sow feed intake was improved slightly. The results from this initial screening study show promise and would warrant further investigation on a larger population of mixed parity, commercially housed sows to access the overall benefits. The benefits of Fysal Fit4 plus above Fysal™ (a currently available product) do however appear marginal and the continued use of Fysal™ would be advocated at this time.

6. Limitations/Risks

The experiment was designed as a screening study to assess potential improvements in lactation outcomes and faecal microbial populations from dietary acidifiers. The study utilized all gilts for two reasons, firstly gilt litters are the most susceptible to disease challenges such as *E.coli* and secondly, at the farm where the study was undertaken the gilts were the only animals that could be hand fed four different diets during late gestation and lactation. The animals were part of a mixed parity herd, and were moved into the farrowing house with older parity sows. Because the sows needed to continue on the four test diets throughout lactation, all of the gilts had to be housed in one area of the farrowing shed to facilitate feeding of the test diets multiple times a day. In a normal production flow environment, producers tend not to house all of the gilts together in the shed due to the higher disease susceptibility of these litters and the ease of transfer of disease from one litter to the next. The outcomes of this current study are likely impacted by this necessary design element and as such, further investigation would be warranted to determine the full effects of feeding Fysal Fit4 Plus in late gestation and lactation to a mixed parity treatment group. Any future study could be narrowed down to only two treatments with the diets fed to all parity sows within the farrowing week.

7. Recommendations

As a result of the outcomes in this study the following recommendations have been made:

- The inclusion of Fysal Fit4 Plus in late gestation and lactation diets reduces key pathogenic microbial populations in lactating sows.
- Numerical improvements in pre-weaning mortality were observed, however further investigation with a larger number population of mixed parity sows would be warranted to confirm and quantify the benefits to producers.
- The benefits of Fysal Fit4 Plus were marginal when compared to Fysal™, and as such the use of Fysal™ given its current availability would be advocated at the current time.

8. References

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