Journal of Applied Microbiology 2005, **99,** 400–407 doi:10.1111/j.1365-2672.2005.02629.x

Impact of dried skim milk in production diets on *Lactobacillus* and pathogenic bacterial shedding in growing-finishing swine[1]

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2004/0952: received 17 August 2004, revised and accepted 8 Decemeber 2004

ABSTRACT

J.E. WELLS, J.T. YEN AND D.N. MILLER. 2005.

Aims: To determine the possible effects of inclusion of dried skim milk (DSM) in swine diets on indigenous *Lactobacillus* spp. and *Escherichia coli*, and its potential for controlling pathogen shedding and affect animal growth in growing-finishing swine.

Methods and Results: Animals were fed over three dietary phases to match production needs from age 10–14 weeks, 14–18 weeks and 18–22 weeks. For each feeding phase, diets were formulated to contain 0 or 10% DSM (balanced for metabolizable energy and true ileal digestible amino acids). Animals were weighed every 2 weeks and faecal samples were collected from 40 animals (20 with DSM and 20 without DSM) at week 10 (d 0 on diets), 14, 18 and 22 of age, and were analysed for *Lactobacillus* spp., Enterobacteriaceae, coliforms, *E. coli*, *Salmonella*, *Campylobacter* and *E. coli* O157:H7. At the start of the study (week 10), faecal bacterial counts ($\log_{10}$ CFU g^{-1} faeces) were 9·55, 7·26, 7·01 and 6·93 for *Lactobacillus*, Enterobacteriaceae, coliforms and *E. coli* populations respectively. The Enterobacteriaceae, coliform and *E. coli* populations decreased through week 14 and 18, but were higher in animals fed with the DSM diet compared with the basal diet without DSM. The *Lactobacillus* populations at weeks 14 and 18 were lower in the animals fed the diet without DSM, whereas feeding DSM maintained the *Lactobacillus* counts from week 10. At week 22, populations of Enterobacteriaceae, coliforms and *E. coli* were >week 18 for the animals fed the diet without DSM, less change was observed with the feeding of DSM, and no differences between the diets were observed at week 22. However, in week 22 the animal gain was positively correlated with *Lactobacillus* numbers and negatively correlated with *E. coli* numbers. Subtraction of the *E. coli* population ($\log_{10}$) from the *Lactobacillus* population ($\log_{10}$) yielded a positive value termed 'effective' *Lactobacillus* that correlated well with animal gain and may better define a beneficial function in the intestine. *Salmonella* were detected in over 60% of the animals at week 10 and 14, and <20% at week 18 and 22. *Campylobacter* were detected rarely at weeks 10, 14 and 18, but were found in 25% of the animals at week 22. The DSM did not affect *Salmonella* or *Campylobacter* shedding, but examination of individual animals over the entire experiment indicated that fewer recurring incidences of *Salmonella* shedding occurred in animals that maintained higher *Lactobacillus*. In addition, at week 22, *Salmonella* and *Campylobacter* shedding was associated with lower levels of effective *Lactobacillus* and lower animal weight gains.

Conclusions: The DSM did not directly affect the animal performance or pathogen shedding via the *Lactobacillus* spp. population at any phase of production. However, analysis of data from all animals revealed that faecal *Lactobacillus* affected *Salmonella* shedding and in the finishing phase, animal growth and pathogen shedding also were affected, as reflected by the 'effective' *Lactobacillus*-associated observations.

[1]*Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the US Department of Agriculture.*

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Significance and Impact of the Study: In the swine intestine, any benefits from gastrointestinal *Lactobacillus* may be compromized by the *E. coli* population, and this antagonism may explain responses observed with prebiotics or probiotics in some swine.

Keywords: *Lactobacillus*, *Escherichia coli*, swine, pathogens, dried skim milk, prebiotic.

INTRODUCTION

Probiotic organisms and prebiotic compounds were used in mammalian diets to alter microbial ecology and generate a healthier gut microflora (Gibson and Roberfroid 1995). Recently, interest in live culture probiotics such as the lactic acid bacteria, has arisen to improve gut performance and reduce pathogen shedding (FAO/WHO 2001; Fernandez *et al.* 2003). In contrast, prebiotics select for beneficial bacteria and/or sustain probiotic strains. The milk sugar lactose has been proposed to have prebiotic properties in certain situations (Szilagyi 2002, 2004), and many gastrointestinal *Lactobacillus* spp. are known to utilize lactose (Kandler and Weiss 1986). Nonfat dried skim milk (DSM) contains 50% lactose (Mahan 2003) and surplus nonfat DSM is often made available from the US Department of Agriculture Commodity Credit Corporation for animal feed and drought assistance, but this feed has not been evaluated for production swine diets in the US.

Pathogen shedding by farm animals is a problem not only for meat producers, but also to the environment and in recent years, pathogen contamination of land, produce and water has become a growing concern in agriculture. Numerous studies have examined pathogens in swine at slaughter and several studies have surveyed pathogen shedding of swine operations. *Salmonella* (Harvey *et al.* 1999a; Barber *et al.* 2002; Bonardi *et al.* 2003; Korsak *et al.* 2003; Kranker *et al.* 2003) and *Campylobacter* (Harvey *et al.* 1999a, 1999b; Young *et al.* 2000) are known zoonotic pathogen inhabitants of the swine gastrointestinal tract. *Escherichia coli* O157:H7 is a recognized problem in animal faeces but is less common in swine faeces, although a prevalence of *E. coli* O157 in 3% of samples has been reported (Bonardi *et al.* 2003). Differences between animal diets and gastrointestinal flora may be a reason for lower prevalence in swine, and in the study by Bonardi *et al.* (2003) swine contaminated with *E. coli* O157 were also fed milk serum, which is a high lactose dairy product. Dietary lactose may be associated with this observation as most *E. coli* bacteria are known to ferment lactose (Brenner 1984).

This study was designed to establish and better understand microbial and pathogen interactions during the process of swine production, and specifically to determine microbial consequences of utilizing DSM as a partial replacement for soya bean meal. The DSM has high amino acid quality and would provide an excellent protein source for commercial swine diets (Pond and Maner 1984). However, changes in dietary components could effect gastrointestinal microbial populations, alter shedding of coliforms and pathogens into the environment and affect animal performance. Fresh faecal samples were taken from animals prior to dietary shifts associated with the swine production system, and populations of *Lactobacillus*, Enterobacteriaceae, coliforms and *E. coli* were enumerated. *Escherichia coli* O157:H7, *Campylobacter* and *Salmonella* were also determined by enrichment and isolation. Potential relationships between microbial flora and animal performance were also examined.

MATERIAL AND METHODS

Animals and design

The experimental protocol (experiment no. 5438-31000-078-02) was approved by the Animal Care and Use Committee of the US Meat Animal Research Center in accordance to the Federation of Animal Science Societies published guidelines (FASS 1999). The swine were a composite offspring of Duroc, Landrace and Large White breeds and were part of larger study (180 animals) conducted to determine the replacement nutritive value of 0, 5 and 10% DSM in growing-finishing diets (Yen *et al.* 2004), but microbial analysis was only conducted with faeces from 40 animals to be fed 0 and 10% DSM. The growing barrows to be fed 0 or 10% DSM (120 castrated males of the 180 total, 8–11 weeks of age; 40·8 ± 2·9 kg body weight) were separated by age into two replicate groups and stratified by weight into outcome groups of 30 animals per treatment (10 pigs per pen and three pens per treatment) within each replicate. To allow for timely collection of the faeces and analysis of the microbial flora, the subset of 40 representative animals (10 animals per treatment within each replicate) with body weights (40·9 ± 0·62 kg body weight) similar to the larger treatment populations (40·8 ± 2·9 kg body weight) were used. Faecal samples were collected from these same 40 animals at the end of each production phase and analysed the same day for microbial pathogens and target populations. Animals were fed in a three-phase

feeding regime (growing, growing–finishing and finishing), typical of many US swine production systems. Each phase diet targets the dietary requirements of the growing–finishing animal and was fed for 4 weeks. Feed and water were provided *ad libitum* in each pen. The pens had slotted floors and were scraped clean regularly. The first-phase basal diet was fed for up to 1 week to animals prior to first sampling and thereafter animals were maintained on treatment diets through each 4-week phase until slaughter. Animals were weighed at the start of study and thereafter every 2 weeks. The animal gain (kg) is reported as the 2-week gain prior to sampling to more closely associate the end of phase microbial flora to the most recent growth. All data are reported on basis of the average age of the animals as they pass through the production phases.

Diets

In a three-phase feeding regimen, corn–soya bean meal basal diets were formulated to match the needs of growing–finishing swine. The 10% DSM was added to the basal diet primarily at the expense of the soya bean meal. Both the basal and 10% DSM diets were formulated to contain the same amounts of calcium, available phosphorus, digestible energy, crude protein and true ileal digestible lysine. To account for changes in the growing–finishing animal's protein requirement, the treatment diets were formulated to provide 15·6%, 13·4% and 11·9% crude protein during phase 1 (growing), phase 2 (growing–finishing) and phase 3 (finishing) respectively. Typical to many US swine production systems, growth-promoting levels of chlortetracycline (0·1% diet) was supplemented to growing phase and growing–finishing phase diets, and bacitracin (0·025% diet) was supplemented to finishing phase diets.

Samples

Faecal samples were collected by rectal massage using a clean sterile glove with each animal. Gloves with sample were inverted and placed into a labelled sealable bag. Following collection, samples were transported to the laboratory and processed on the same day (typically <2 h) for microbial counts (CFU) of *Lactobacillus*, Enterobacteriaceae, coliforms and *E. coli*. Faecal samples were also processed for *E. coli* O157:H7, *Campylobacter* and *Salmonella*.

Microbial analysis

A 5-g portion of each faecal sample was transferred into a Stomacher bag with 45 ml of buffered peptone water (BPW) and gently mixed with a Stomacher (Seward Limited, London, UK) for 30 s. An aliquot (representing a 10^{-1} dilution) was taken from the BPW faecal sample and used for 10-fold serial dilutions to 10^{-6} in a 96-well, deep-dish plates using BPW. Dilutions of 10^{-3}, 10^{-4} and 10^{-5} were transferred to Petrifilms (3 M Microbiology Products, St Paul, MN) to determine CFU of Enterobacteriaceae, coliforms and *E. coli* and 100 μl of 10^{-6} was spread over Modified MRS (pH 5·4 with acetic acid) agar plates to determine CFU of *Lactobacillus* (Krause *et al.* 1995; Wells *et al.* 1997). Petrifilms and plates were incubated overnight at 37°C and CFU was counted. Remaining BPW-faecal sample was incubated for 7 h at 37°C to enrich for *E. coli* O157:H7 and subsequently a 250 μl aliquot was diluted to 1 ml with a phosphate buffer solution for immunomagnetic separation of *E. coli* O157. The Dynabeads anti-*E. coli* O157 beads (Dynal Biotech, Oslo, Norway) were washed per instructions 2–3 times to remove debris and suspended in 100 μl phosphate buffered saline. A 50 μl portion was plated onto MacConkey sorbitol Agar (SMAC) with cefixime-tellurite (CT) supplement (Dynal Biotech) and incubated overnight at 37°C. White colonies on CT-SMAC were tested for agglutination using anti-*E. coli* O157 antibodies (*E. coli* O157 Test Kit, Oxoid Ltd, Hampshire, UK).

Salmonella were enriched from 1 g of faeces with 13-ml tetrathionate Broth in a sterile 15-ml loosely-capped conical tube. Tubes were incubated for 48 h at 37°C and a 10 μl aliquot was transferred to 10-ml Rappaport–Vassiliadis Soya Peptone Broth (RVS; Oxoid Ltd, Hampshire, UK) tube. The RVS tubes were incubated at 42°C for 24 h and then 20 μl was plated onto hektoen-enteric (HE) agar. Black colonies were picked and plated onto fresh HE plate. Isolated colonies were grown in Tryptic Soy Broth and verified to be presumptive *Salmonella* with Triple Sugar Iron Agar and Lysine Iron Agar slants.

Campylobacter was enriched from 1 g of faeces with 13-ml Bolton Broth with supplement (Oxoid) and Lysed Horse Blood Cells (Lampire Biological Labs, Pipersville, PA). Tubes were gently mixed, capped tightly and incubated for 4 h at 37°C followed by 44 h at 42°C. A 10 μl aliquot was plated onto Campy-Cephex agar (Stern *et al.* 1992) and grown using MicroAero Packs in AnaeroPack System (Mitsubishi Gas Chemical, New York, NY) for 48 h at 42°C. Colonies were verified presumptive by agglutination (*Campylobacter* Test Kit, Oxoid).

Chemicals and reagents were from Sigma (St Louis, MO) unless otherwise noted. Bacterial media and agars were DIFCO brand (B-D Microbiological Systems, Sparks, MD) unless otherwise noted. Microbial numbers were transformed to $\log_{10}$ and data sets were statistically analysed using Student's t-test for significant differences (Sokal and Rohlf 1969). Linear and curve-linear relationships were fit using KaleidaGraph data analysis and graphing software package (Synergy Software, Reading, PA).

RESULTS

The 10-week-old barrows had 9·55, 7·26, 7·01 and 6·93 $\log_{10}$ CFU g^{-1} faeces for *Lactobacillus*, Enterobacteriaceae, coliforms, and *E. coli* populations in their faeces, respectively, prior to dietary treatments. By the end of the growing phase (age 14-week), the *Lactobacillus* counts in basal fed animals had decreased nearly 0·25 $\log_{10}$ (Table 1), whereas no decrease was observed in this population when the animals were fed 10% DSM. Bacterial counts of Enterobacteriaceae, coliforms and *E. coli* for both treatments were lower, but animals on the 10% DSM diet had significantly higher levels of each population relative to the basal fed animals.

At the end of the growing-finishing phase (age 18-week), the *Lactobacillus* counts for both treatments were similar to those observed at the end of the growing phase (Table 1). Although the Enterobacteriaceae, coliforms and *E. coli* counts were much lower relative to the previous growing phase, these counts were still significantly higher with the 10% DSM diet. Following the finishing phase (age 22-week), the *Lactobacillus* counts were lower in both treatments relative to the previous phases (Table 1), but the decrease was greater for the 10% DSM diet and little difference was observed between the two treatments. The counts for Enterobacteriaceae, coliforms and *E. coli* increased for both treatments relative to the counts observed in the previous phase, but more so for animals consuming the basal fed diet.

Pathogen shedding was similar for both treatments (Table 1), but differed according to the production phase. Presumptive *Salmonella* were detected in 62% of the animals at the beginning of the experiment (age 10-week) and at least once in all but three animals (7·5% negative) tested in this study. *Salmonella* shedding was greatest in both basal and 10% DSM fed animals following the growing phase, and lowest following the growing-finishing phase (Table 1). *Campylobacter* shedding was observed in only 5% of the animals prior to the dietary treatments and in 32% of the animals over the experiment. During the production phases as sampled, a significant level of *Campylobacter* shedding was predominantly observed following the finishing phase (Table 1). In the finishing animals, the percent *Campylobacter* shedding was similar to the percent *Salmonella* shedding, but only three of the 14 pathogen-shedding animals were shedding both *Salmonella* and *Campylobacter* at this time. *Escherichia coli* O157 was not detected in faeces from any sample collected in this study. No extraordinary difference was observed between the basal and 10% DSM diets in pathogen shedding (Table 1), but closer examination of the collected data set suggested that persistent shedders of *Salmonella* (grouped by the total number of positives for each animal over the four samplings) maintained lower average *Lactobacillus* numbers throughout the study (Fig. 1).

Measures of animal productivity (kg gained per day or total kg gain) over the entire study were not correlated with average faecal *Lactobacillus*, Enterobacteriaceae, coliforms or *E. coli* numbers (data not given). However, higher productivity (prior 2-week kg gain, age 20–22-week, both dietary

Table 1 The effect of dried skim milk in production swine diets on the faecal commensal flora and pathogenic bacterial shedding*

Period	Pretrial	Growing phase (week 4)			Growing-finishing phase (week 8)			Finishing phase (week 12)		
Pig age	10 week	14 weeks			18 weeks			22 weeks		
		Basal diet	10% DSM	*P*-value	Basal diet	10% DSM	*P*-value	Basal diet	10% DSM	*P*-value
Bacterial population†										
Commensals ($\log_{10}$ g^{-1} faeces)										
Lactobacillus spp.	9·55 ± 0·05	9·29 ± 0·08	9·59 ± 0·06	<0·01	9·35 ± 0·12	9·54 ± 0·07	<0·1	9·19 ± 0·14	9·06 ± 0·15	<0·5
Enterobacteriaceae	7·26 ± 0·15	5·65 ± 0·14	6·56 ± 0·15	<0·01	4·90 ± 0·16	5·48 ± 0·22	<0·05	5·71 ± 0·21	6·01 ± 0·25	<0·5
Coliforms	7·01 ± 0·15	5·58 ± 0·12	6·32 ± 0·15	<0·01	4·74 ± 0·17	5·46 ± 0·24	<0·05	5·49 ± 0·23	5·61 ± 0·22	<0·9
E. coli	6·93 ± 0·15	5·52 ± 0·13	6·31 ± 0·15	<0·01	4·67 ± 0·16	5·41 ± 0·24	<0·05	5·32 ± 0·25	5·36 ± 0·24	<0·9
Pathogens (% of animals)										
Salmonella	62	60	65		10	15		15	25	
Campylobacter	5	5	0		0	0		20	30	
E. coli O157	0	0	0		0	0		0	0	

Growing phase = 15·6% crude protein.

Growing-finishing phase = 13·4% crude protein.

Finishing phase = 11·9% crude protein.

Chlortetracycline was included in Growing and Growing-finishing diets, and bacitracin was included in the Finishing diet.

*Animals were sampled pre-trial at 10 weeks of age and at the end of each production phase (4-week period). Dietary protein was balanced for each based on the animal's requirement.

†Bacterial populations are averages of 20 animals per treatment group.

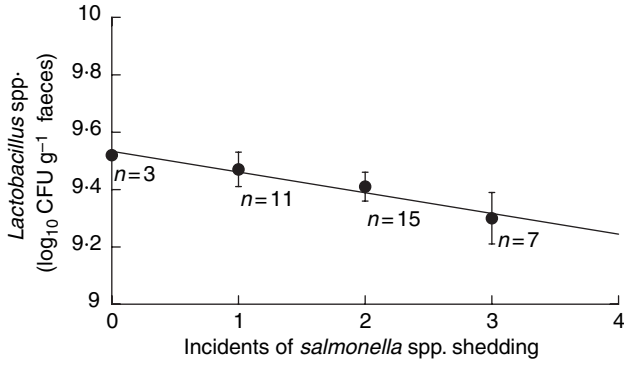

Fig. 1 Relationship between incidents of *Salmonella* shedding by individual animals and the average number of faecal *Lactobacillus* counts in those animals over the four samplings ($R^2 = 0{\cdot}92$). Complete data from all samplings was obtained for 36 of the 40 animals in the study, and the animals were grouped by total incidents of shedding over the four samplings. The number of animals (n) within each shedding group is noted with respective data point in the figure

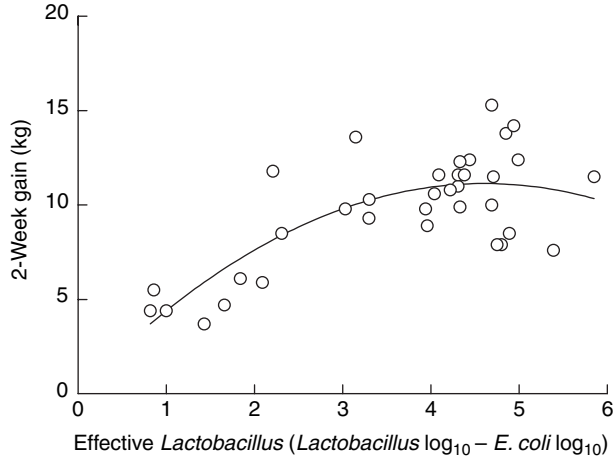

Fig. 3 Relationship between faecal 'effective' *Lactobacillus* (*Lactobacillus* $\log_{10}$ week 22 counts – *Escherichia coli* $\log_{10}$ week 22 counts) following the finishing phase and each animal's prior 2-week kg gain (from age 20-week to 22-week)

treatments) was associated with higher *Lactobacillus* numbers ($R^2 = 0{\cdot}33$) and lower *E. coli* numbers ($R^2 = 0{\cdot}35$) in the animal's faeces at the end of the finishing phase (age 22-week, Fig. 2). No correlation between *Lactobacillus* and *E. coli* numbers was observed ($R^2 < 0{\cdot}1$) between these same animals, but a simple subtraction of the *E. coli* population from the *Lactobacillus* population resulted in 'effective' *Lactobacillus* and this number correlated well ($R^2 = 0{\cdot}48$ linear, $R^2 = 0{\cdot}58$ curve-linear) with these animals prior 2-week gain at the end of the finishing phase (Fig. 3). During the finishing period, most poor production was associated with animals that had $<2{\cdot}5$-$\log_{10}$ differences between *Lactobacillus* and *E. coli* populations. Similar analysis of the previous phases did not reveal the same type of relationship between prior 2-week performance and microbial populations, but in these earlier growing and growing-finishing phases, few animals performed poorly and almost all animals sustained $>2{\cdot}5$-$\log_{10}$ differences between the microbial populations (data not given).

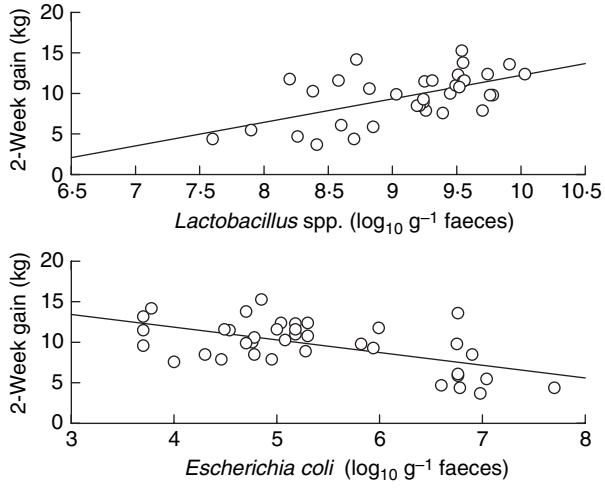

Fig. 2 Relationship between *Lactobacillus* [(a) $R^2 = 0{\cdot}32$, linear] and *Escherichia coli* [(b) $R^2 = 0{\cdot}34$, linear] counts in faeces collected following the finishing phase (age 22-week) and each animal's prior 2-week kg gain (from age 20-week to 22-week)

At the end of the growing phase, *Salmonella* was the predominant pathogen shed in the faeces (Table 1) and the 62% animals shedding resulted in slightly lower animal kg gains ($12{\cdot}9 \pm 0{\cdot}33$ *vs* $13{\cdot}8 \pm 0{\cdot}56$, $P = 0{\cdot}15$). At the end of the finishing phase, *Salmonella* and/or *Campylobacter* were shed in the faeces of 14 animals and they also tended to have lower animal kg gains ($8{\cdot}3 \pm 1{\cdot}10$ *vs* $10{\cdot}0 \pm 0{\cdot}54$, $P = 0{\cdot}14$). In the finishing phase, the shedding of pathogens appeared to be negatively associated with faecal *Lactobacillus* numbers (data not given), but a stronger relationship was observed between pathogens and 'effective' *Lactobacillus* (Fig. 4). Animals with the highest effective *Lactobacillus* at the end of the finishing phase (age 22-week) gained better in the 2 weeks (age 20-week to 22-week) prior to sampling (Fig. 4a), and fewer of these animals were found to shed pathogens in the faeces at the end of the finishing phase (Fig. 4b).

DISCUSSION

Lactobacillus spp. are important inhabitants of the mammalian gastrointestinal system and much work has emphasized increasing their numbers to improve gastrointestinal health (Gibson and Roberfroid 1995; FAO/WHO 2001). In swine,

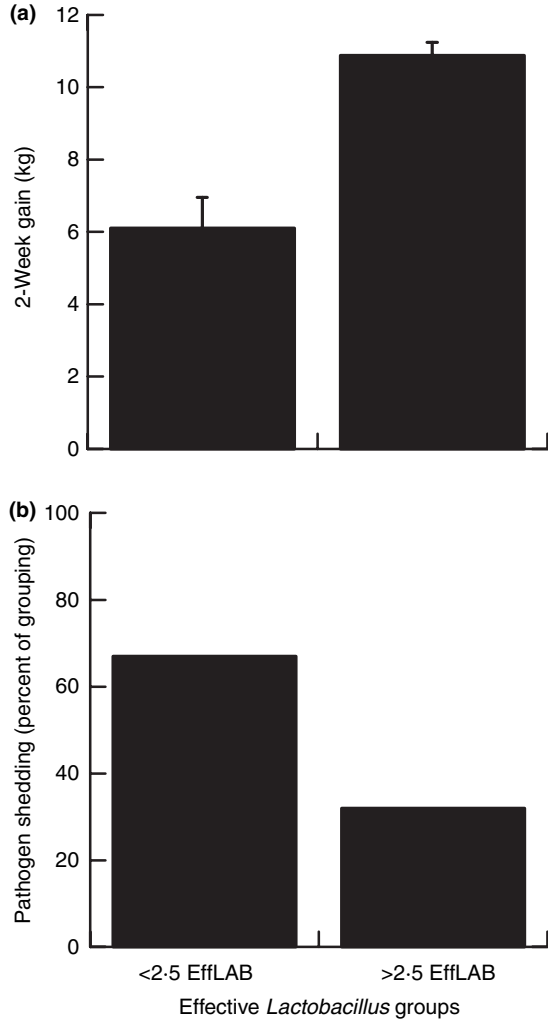

Fig. 4 Average kg gain (from week 20–22) and percentage of animals shedding pathogens (at week 22). Animals from both treatments were sorted into two groups based on 'effective *Lactobacillus*' [above ($n = 26$) or below ($n = 9$) 2·5 $\log_{10}$ difference] at week 22

the benefits of lactose in the weaning pig diet was demonstrated by Mahan (2003), but little work was performed with older animals. Lactose has been proposed to be a prebiotic when host lactase activity is limited (Szilagyi 2002, 2004) and in swine, the lactase activity is greatest after birth, but considerably decreases after 2 weeks of age (Yen 2001). The DSM has *c.* 50% lactose (Mahan 2003) and is an excellent dietary nutrient with high amino acid quality (Pond and Maner 1984). In the US, DSM remains a dairy commodity with significant federal purchases to sustain milk and dairy prices for producers and as a consequence, surplus DSM is stored and made available for nonhuman use.

In this study, DSM did sustain higher levels of *Lactobacillus* in the animal faeces following growing and growing–finishing phases of production, and would appear to act, by this indicator, as a prebiotic in swine diets under certain conditions. The addition of DSM increased the total numbers (or biomass) of *Lactobacillus* nearly by twofold ($1{\cdot}95 \times 10^9$ *vs* $3{\cdot}89 \times 10^9$ CFU), but no associated benefit to animal performance (kg gain) or decrease in pathogen shedding was observed with increased *Lactobacillus*. The dietary DSM also yielded higher Enterobacteriaceae, coliform and *E. coli* counts, but most of the Enterobacteriaceae and coliform counts were *E. coli*. As *E. coli* can utilize lactose as an energy and carbon source, the possibility that the increased *E. coli* limited or negated the potential benefits from higher *Lactobacillus* with DSM addition needs to be further studied in growing and growing–finishing swine.

The *E. coli* and coliform counts decreased nearly 2·3 $\log_{10}$ in the 8-week period from pretrial to the end of the growing–finishing phase for the basal fed animals. A smaller, but continuous decrease was also observed over this same time with DSM fed animals. These decreases may reflect the adaptation of the growing animal's digestive system, gradual effects of low levels of the growth promoting antibiotic chlortetracycline, and/or decreases in dietary protein on the gastrointestinal flora. These decreasing microbial numbers were not anticipated in our experimental design and need future studies to better assess the changes in microbial flora associated with production processes and to determine their impact on animal production.

Following the finishing phase, we observed a positive relationship between faecal *Lactobacillus* and the prior 2-week gain for individual animals, and we also observed a negative relationship between faecal *E. coli* and 2-week gain. This later observation with indigenous, nonpathogenic *E. coli* has not been previously reported. A recent study did report lower Enterobacteriaceae when the probiotic *Lactobacillus murinus* was fed to swine (Gardiner *et al.* 2004), but measures of animal performance were not reported. In our study, *Lactobacillus* did not negatively correlate with the *E. coli* populations, but a different relationship may exist between these populations and the host. The faecal *Lactobacillus* population was always greater than *E. coli* population, and the difference ('effective' *Lactobacillus*) between the $\log_{10}$ number of *Lactobacillus* and the $\log_{10}$ number of *E. coli* correlated well with the animal's prior 2-week gain during the finishing phase (Fig. 3). Specifically, animals with less than a 2·5-$\log_{10}$ difference had the least amount of gain.

Pathogen shedding varied over the production phases but was not significantly affected by DSM. More important, *E. coli* O157:H7 was not detected in the faecal samples although the DSM animals had much higher numbers of *E. coli* in general. *Salmonella* are commonly associated with swine production (Harvey *et al.* 1999a; Barber *et al.* 2002; Bonardi *et al.* 2003; Korsak *et al.* 2003; Kranker *et al.* 2003).

In this study, *Salmonella* shedding was greatest after the growth phase (week 4), and those animals shedding *Salmonella* had only slightly lower gains (6·5% lower). Few *Salmonella* were found in the faeces following the growing-finishing phase, but after the finishing phase, the number of animals shedding *Salmonella* nearly doubled and a similar number of animals was also found to be shedding *Campylobacter*. Animals shedding pathogens during the finishing period tended to have less 2-week weight gain (16% lower) prior to faecal sampling and lower effective *Lactobacillus* (Fig. 4). This relationship between productivity, microbial flora and pathogen shedding will need to be studied in greater detail with particular attention to antibiotics in the swine rations. Chlortetracycline and bacitracin are typically added to diets as a growth promoter in many swine production systems, and the microbial flora in our study may not have recovered from the chlortetracycline to bacitracin change between the growing-finishing phase and finishing phase diets. Nonetheless, the effective *Lactobacillus* appears to indicate a healthier microbial population in the gastrointestinal system.

Lactobacillus was proposed to decrease pathogen shedding (FAO/WHO 2001; Fernandez *et al.* 2003) and probiotics was fed to decrease pathogens in the faeces (Harvey *et al.* 2002; Nisbet 2002; Genovese *et al.* 2003). In this study, we did not observe a relationship between the pathogen shedding and faecal *Lactobacillus* at any time point in the study. However, we analysed the data for all animals over the entire time and observed a linear response between *Salmonella* shedding and average number of *Lactobacillus* in the faeces of those animals (Fig. 1). Animals that sustained higher *Lactobacillus* numbers throughout the production phases shed fewer *Salmonella*, and this observation suggests a long-term role for *Lactobacillus* in maintaining a healthy gastrointestinal system. It would appear that maintaining higher *Lactobacillus* could control long-term pathogen shedding from production animals and agrees with many of the probiotic strategies as preventative measures.

The use of DSM in swine diets did not alter animal performance, meat quality or nitrogen excretion (Yen *et al.* 2004) and based on the observations reported here, DSM does not significantly alter pathogen shedding in faeces when included in typical production diets. The potential for *Lactobacillus* to affect animal gain and pathogen shedding was noted in individual animals under certain conditions, but prebiotic effects of DSM in our animals may have been limited because of the antibiotics included in animal rations. These compounds are currently used in production systems as growth promoters and therefore were included in this experiment, but future research will be needed to determine if DSM provides beneficial effects in the absence of antibiotics. Nonetheless, utilization of DSM in production diets would be a viable alternative for surplus or outdated DSM, but may not significantly affect pathogen shedding from animals.

ACKNOWLEDGEMENTS

The authors thank C.L. Felber and J.M. Long for technical assistance, T. Acton and associates for animal management, and J.K. Byrkit for secretarial assistance.

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