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INFLUENCE OF A PROBIOTIC/TRACE MINERAL MIXTURE ON GROWTH PERFORMANCE AND *SALMONELLA CHOLERAESUIS* SHEDDING IN NURSERY PIGS¹

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Summary

We tested a probiotic/trace mineral mixture using a bacterial challenge model in high-health status pigs. We examined the influence of the mixture on growth performance, hematologic parameters, haptoglobin concentration, and *Salmonella choleraesuis* shedding in nursery pigs. A successful model of *S. choleraesuis* challenge was established. However, the probiotic/trace mineral mixture did not influence growth performance, bacterial shedding, or other parameters examined in this experiment.

(Key Words: *Salmonella choleraesuis*, Probiotics, Trace Minerals.)

Introduction

The influences of disease processes on growth performance are difficult to quantify in actual on-farm conditions. Therefore, disease challenge models are employed in carefully controlled environments to quantify the influence of therapeutic agents.

In addition to growth performance benefits, therapeutic agents can be used to decrease the bacterial numbers shed or residing in the gastrointestinal tract. The decreased numbers then result in less contamination of meat products in the packing plant. Currently, probiotics are used in Sweden to reduce *Salmonella* bacterial shedding in chickens and are being tested in cattle to reduce *E. coli* shedding.

Consequently, our objective was to use a bacterial challenge model to determine the influence of a probiotic trace mineral mixture on growth performance, hematologic parameters, haptoglobin concentration, and *Salmonella choleraesuis* shedding in nursery pigs.

Procedures

Sixty-four high-health status pigs (initially 28.7 lb, PIC L326 sires × C15 dams) were obtained from a commercial farm in north-east Kansas and used in three experiments. Neither clinical signs nor laboratory evidence of *S. choleraesuis* infection had been observed on this farm. In addition, neither clinical signs nor laboratory evidence of any other enteric diseases had been observed in the group of pigs from which the experimental pigs were obtained. Pigs were housed in an environmentally controlled isolation facility in which the initial temperature (75°F) was reduced by 2°F each week. Each pen contained a self-feeder and a nipple waterer to provide ad libitum access to feed and water.

Experiment 1. Experiment 1 was a 35-day assay. Pigs were allotted by initial weight, with gender equalized across treatments within blocks to polyethylene pens (4 × 5 ft) with slotted plastic flooring, using four pigs per pen and eight replicate pens.

Pigs were fed either a control diet or a diet containing probiotic/trace minerals for the 35 d trial (Table 1). All diets were

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formulated to contain 1.25% lysine, .345% methionine, .80% Ca, and .70% P. On day 14, all pigs were challenged by oral gavage with 8.7×10^9 cfu of *S. choleraesuis* bacteria after being held off-feed for 6 hours. The *S. choleraesuis* isolate was obtained from the liver and spleen of a pig submitted to the Kansas Veterinary Diagnostic Laboratory. The challenge dose had been determined to decrease growth rate by approximately 16% and induce clinical signs of *S. choleraesuis* infection in a previous pilot study. Prior to challenge, fecal swabs were taken from one pig per pen and cultured for *S. choleraesuis*; no *S. choleraesuis* was isolated. Fecal swabs were obtained from all pigs on d 24 and 31 and cultured for *S. choleraesuis*.

Pig weights and feed consumption were determined weekly to calculate ADG, ADFI, and F/G. On d 17, 24, and 31, serum and whole blood samples were collected from each pig and analyzed for haptoglobin, white blood cell, red blood cell, hemoglobin, and mean corpuscular hemoglobin (MCHC) concentrations, as well as hemotocrit percentage, mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH).

Experiment 2. Twenty-eight of the pigs fed the control diets in experiment 1 were reallocated by weight, with gender equalized across treatments within blocks to polyethylene pens (4 × 5 ft) with slotted plastic flooring, using two pigs per pen and seven replicate pens. Experiment 2 was an 18-day growth assay. Pigs were housed in an environmentally controlled isolation facility in which the temperature was maintained at 65°F. Each pen contained a self-feeder and a nipple waterer to provide ad libitum access to feed and water.

Pigs were fed either the control diet or a diet containing probiotic/trace minerals; both contained carbadox (25 g/ton; Table 1). An antibiotic sensitivity had been performed previously and indicated that the *S. choleraesuis* isolate used in this experiment was sensitive to carbadox. All diets were formulated to contain 1.25% lysine, .345% methionine, .80% Ca, and .70% P.

Pig weights and feed consumption were determined to calculate ADG, ADFI, and F/G.

Experiment 3. Twenty-eight of the pigs fed the diets containing the probiotic/trace mineral mixture in Experiment 1 were reallocated by weight, with gender equalized across treatments within blocks to solid-floor concrete pens (6 × 8 ft) with solid concrete pen dividers, using three or four pigs per pen and four replicate pens. Pigs were housed in an isolation facility in which a thermoneutral environment was maintained. Experiment 3 was an 18-day growth assay.

Pigs were fed either a control diet or a diet containing carbadox (25 g/ton). Both diets contained the probiotic/trace mineral mixture (Table 1). Both diets were formulated to contain 1.25% lysine, .345% methionine, .80% Ca, and .70% P.

Pig weights and feed consumption were determined to calculate ADG, ADFI, and F/G.

Statistical Analysis. Data were analyzed according to the GLM procedures of SAS as a randomized complete block design with initial body weight used to establish each block. Haptoglobin concentrations and hematology parameters were analyzed using a repeated measures ANOVA. Pen was used as the experimental unit for all statistical analysis.

Results and Discussion

Experiment 1. During the d 0 to 14 period, no differences were detected in growth performance (Table 2). One pig died during this period. Subsequent bacteriological and histological examinations indicated that the pig died from a bacterial meningitis caused by *Streptococcus suis*.

The *Salmonella* challenge model used in this experiment was successful in establishing clinical signs and fecal shedding of *S. choleraesuis* (Table 3). However, no differences in the prevalence of shedding occurred between the two treatment groups. Further

evidence of clinical disease was the decreased growth performance from d 14 to 35 of the experiment. However, again no differences occurred between treatments.

Three pigs (two on the control diet and one on the probiotic/trace mineral diet) died during the day 14 to 35 period. Subsequent bacteriological and histological examinations indicated that they died from a bacterial septicemia caused by *S. choleraesuis*.

Haptoglobin and white blood cell concentrations were elevated in both treatment groups, with no difference between groups (normal values are < 15 mg/dL and 11 to 22 × 10³/μL for haptoglobin and white blood cells, respectively; Table 4). Elevated haptoglobin and white blood cell count are

indicative of an infectious disease. All other hematology parameters were within normal limits, with no differences between treatments.

Experiments 2 and 3. Growth performance was excellent in both experiments (Tables 5 and 6). However, neither the probiotic/trace mineral nor carbadox had an influence on growth performance. One pig in the carbadox group in experiment 3 died on d 17 of the experiment.

In conclusion, a successful model of *S. choleraesuis* challenge was established. However, probiotic/trace mineral mixture did not influence growth performance, bacterial shedding, or other parameters examined in this experiment.

Table 1. Diet Composition, (%) As-Fed

Ingredient, lb	Experiment 1		Experiment 2		Experiment 3	
	Control	PBTM ^a	Carbadox		PBTM	
			Control	PBTM	Control	Carbadox
Corn	60.02	60.02	59.52	59.52	59.52	59.52
Soybean meal (46.5% CP)	32.53	32.53	32.53	32.53	32.53	32.53
Soy oil	3.00	3.00	3.00	3.00	3.00	3.00
Monocal (21% P)	1.53	1.53	1.53	1.53	1.53	1.53
Limestone	1.09	1.09	1.09	1.09	1.09	1.09
Salt	.35	.35	.35	.35	.35	.35
KSU swine vitamin premix	.25	.25	.25	.25	.25	.25
KSU swine trace mineral	.15	.15	.15	.15	.15	.15
Lysine	.15	.15	.15	.15	.15	.15
Methionine	.025	.025	.025	.025	.025	.025
Medication ^b	--	--	.50	.50	--	.50
Belly Buster	--	.50	--	.50	.50	.50
Iron Vite	--	.40	--	.40	.40	.40
Corn Starch	.90	--	.90	--	.50	--
Total	100.00	100.00	100.00	100.00	100.00	100.00
		2000.00	2000.00	2000.00	000.00	2000.00

^aPBTM = Probiotic/trace mineral mixture (Belly Buster P and Iron Vite, Porter Livestock Products, Nichols, IA.

^bProvided 25 g/ton carbadox (Mecadox, Pfizer).

Table 2. Experiment 1 Growth Performance^a

Item	Control 1	PBTM	CV
<u>d 0 to 14</u>			
ADG, lb	.92	.82	12.0
ADFI, lb	1.90	1.84	6.7
F/G	2.14	2.24	15.2
<u>d 14 to 35</u>			
ADG, lb	1.07	1.00	19.7
ADFI, lb	2.23	2.12	8.7
F/G	2.09	2.25	20.3
<u>d 0 to 35</u>			
ADG, lb	1.01	.93	11.2
ADFI, lb	2.09	2.00	5.6
F/G	2.09	2.20	8.0
<u>d 14 to 21</u>			
ADG, lb	.34	.41	134.5
ADFI, lb	1.24	1.38	17.4
F/G	3.84	4.25	161.7

^aEach number is the mean of eight replicate pens (four pigs per pen with a mean initial weight of 28.7 lb). Pigs were challenged orally with 8.7×10^9 cfu of *Salmonella choleraesuis* bacteria 14 days after the beginning of the test period. Pigs were fed the diets for 35 days. No significant differences were detected ($P > .15$).

Table 3. Recovery of *Salmonella choleraesuis* from Fecal Swabs in Experiment 1^a

Culture Date	Control 1	PBTM
Day 24	7/31 (23%) ^b	8/31 (26%)
Day 31	13/31 (42%)	10/30 (33%)

^aPigs were challenged orally with 8.7×10^9 cfu of *Salmonella choleraesuis* bacteria 14 days after the beginning of the test period.

^bNumber pigs positive / Number of pigs cultured (Percentage).

Table 4. Haptoglobin Concentration and Hematology Parameters in Experiment 1

Item	Control 1	PBTM	<i>P</i> <	CV
Haptoglobin, g/dL	97.3	91.9	.32	26.6
White blood cell count, $\times 10^3/\mu\text{L}$	30.2	29.9	.83	14.3
Red blood cell count, $\times 10^6/\mu\text{L}$	7.6	6.3	.24	50.7
Hemoglobin, g/dL	11.4	10.9	.09	3.1
Hematocrit, %	34.2	32.8	.10	3.5
MCV, fL	52.4	52.2	.70	1.4
MCH, pg	17.4	17.3	.65	1.4
MCHC, g/dL	32.5	33.1	.41	7.3

^aEach number is the mean of eight replicate pens (four pigs per pen with a mean initial weight of 28.7 lb). Pigs were challenged orally with 8.7×10^9 cfu of *Salmonella choleraesuis* bacteria 14 days after the beginning of the test period. Pigs were fed the diets for 35 days.

Table 5. Experiment 2 Growth Performance (Pigs Fed the Control Diet in Experiment 1)^a

Item	Carbadox		CV
	Control 2	PBTM	
ADG, lb	1.80	1.75	6.1
ADFI, lb	3.77	3.67	11.3
F/G	2.08	2.10	7.9

^aEach number is the mean of seven replicate pens (two pigs per pen with a mean initial weight of 62.1 lb). Pigs were challenged orally with 8.7×10^9 cfu of *Salmonella choleraesuis* bacteria 21 days before the beginning of the test period. Pigs were fed the diets for 18 days. No significant differences were detected ($P > .15$).

Table 6. Experiment 3 Growth Performance (Pigs Fed the Probiotic/Trace Mineral Mixture in Experiment 1)^a

Item	PBTM		CV
	Control 3	Carbadox	
ADG, lb	1.85	1.75	4.4
ADFI, lb	3.63	3.56	4.2
F/G	1.97	2.05	6.9

^aEach number is the mean of four replicate pens (three or four pigs per pen with a mean initial weight of 62.5 lb). Pigs were challenged orally with 8.7×10^9 cfu of *Salmonella choleraesuis* bacteria 21 days before the beginning of the test period. Pigs were fed the diets for 18 days. One pig from the carbadox group died on d 17. No significant differences were detected ($P > .20$).