# Effect of a *Bacillus*-based direct-fed microbial feed supplement on growth performance and pen cleaning characteristics of growing-finishing pigs

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**ABSTRACT:** A direct-fed microbial (DFM) based on a combination of Bacillus organisms specifically selected to increase the manure decomposition process was evaluated to determine its efficacy for improving growth performance and manure dissolution time. Three experiments involving 336 crossbred barrows and gilts were conducted to determine the effect of the Bacillus-based direct-fed microbial on growth performance and pen cleaning time. In each experiment, 2 dietary treatments (0 and 0.05% DFM) were fed during the growing-finishing period throughout the experiment, such that the DFM provided  $1.47 \times 10^8$  cfu of Bacillus organisms per gram of supplement. Data from the 3 experiments were combined for analysis, such that there were 28 pens representing each of the 2 treatments. Pigs were weighed and feed intake was determined at the initiation and termination of each phase (starter, grower, and finisher). At the end of Exp. 1 and 3, pen cleaning time was determined by measuring the time required for each pen to be scraped and washed with a high-pressure sprayer. Additionally, 2 solid manure mat samples weighing approximately 4 g each were collected from solid manure buildup in

each pen (16 pens/treatment), and the time required to completely disperse each manure mat sample was determined. Gain: feed improved (P < 0.05) in pigs fed *Bacillus* compared with the control diet during the finisher phase and throughout the combined growing-finishing period. The time required to dissolve the manure mat was reduced (P < 0.01) in samples collected from pens containing pigs fed Bacillus compared with samples from control pens. An additional evaluation was conducted in a commercial swine production facility using statistical process control analysis. Statistical process control analysis determined that supplementation with Bacillus increased the expected mean for ADG and decreased the expected mean for death loss percentage. Supplementation with a DFM composed of specifically selected Bacillus organisms improved G:F and decreased the time required to disperse a swine manure mat sample in a controlled study conducted at swine research facilities. Furthermore, when evaluated in a commercial swine production facility, the Bacillusbased DFM improved ADG and reduced mortality of pigs during the growing-finishing period.

**Key words:** feed additive, growth, manure, probiotics, swine

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#### INTRODUCTION

Intensified swine production faces many challenges related to managing manure output, such as storage and handling of manure, ammonia and odor production, and the accumulation of solids on pen floor surfaces

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and within manure storage facilities. Odor production and accumulation of manure solids are characteristics that manifest as a result of inadequate microbial decomposition of manure. This is further compounded by swine diet formulations, which commonly contain high concentrations of trace minerals and antibiotics that have deleterious effects on the bacteria needed for effective manure decomposition (Gilley et al., 2000). Many solutions have been devised to alleviate problems associated with odor and manure handling, including aeration of stored manure, diet manipulation through addition of crystalline amino acids or fermentable carbohydrates, enzyme and microbial additives, and odormasking compounds (Sutton et al., 1999). However,

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these methods are expensive, inconvenient, or do not adequately support the manure decomposition process.

Bacillus species are ideally suited as feed additives due to their stability as spore-forming bacteria and ability to produce a variety of enzymes to promote manure digestion (Schreier, 1993). Because Bacillus are capable of producing spore coats that afford protection from heat, enzymatic degradation, and the acidic conditions of the stomach (Hong et al., 2005), dietary supplementation with manure-degrading microorganisms would provide a convenient and continuous inoculation strategy for manure storage facilities.

This study was conducted to evaluate the ability of dietary supplementation with specifically selected strains of *Bacillus subtilis* and *Bacillus lichenformis* to improve pen cleaning and growth performance of growing/finishing pigs. Furthermore, the effect of these *Bacillus* strains on growth performance was evaluated in commercial swine production facilities using statistical process control (**SPC**) analysis.

#### MATERIALS AND METHODS

# University Research Experiment

Animal management and experimental procedures conducted during this study were approved by the University of Arkansas Institutional Animal Care and Use Committee.

Three experiments were conducted utilizing a total of 336 crossbred barrows and gilts (Hampshire × Duroc sires mated to crossbred sows) at the University of Arkansas Swine Research Unit. At the commencement of each study, pigs were moved from nursery facilities to a growing/finishing barn located at the same farm site, blocked by initial BW ( $24.2 \pm 1.4 \text{ kg}$ ,  $27.0 \pm 1.4 \text{ kg}$ , and  $21.2 \pm 1.1$  kg for Exp. 1, 2, and 3, respectively), and housed in pens  $(1.5 \times 4.0 \text{ m})$  holding 6 pigs each (a total of 16 pens each in Exp. 1 and 2, and 24 pens in Exp. 3). The growing/finishing facility was approximately 30 vr old and consisted of a naturally ventilated curtainsided barn. The first study was conducted from February to April, the second study from July to September, and the third study from June to August. Pens contained partially slatted concrete flooring, in which the solid portion of the pen measured  $2.8 \times 1.5$  m and the slatted portion measured  $1.2 \times 1.5$  m.

Pigs were blocked by initial BW, such that there were 8 BW blocks in Exp. 1 and 2 and 3 BW blocks in Exp. 3. Pigs were randomly allotted within BW block to 1 of 2 dietary treatments (as-fed basis): a control basal diet (Table 1) or the control diet supplemented with 0.05% of a Bacillus-based direct-fed microbial (MicroSource "S," Agtech Products Inc.) added at the expense of corn on an equal weight basis. The direct-fed microbial delivered  $1.47 \times 10^8$  cfu of Bacillus organisms comprising 2 strains of B. lichenformis and 1 strain of B. subtilis per gram of supplement. Basal diets were formulated to meet or exceed the nutrient requirement recommenda-

**Table 1.** Composition of basal diets fed to growing-finishing pigs on an as-fed basis during the university research study<sup>1</sup>

Item	Starter <sup>2</sup>	$Grower^2$	Finisher <sup>2</sup>
Ingredient, %			
Yellow corn	62.615	64.495	68.785
Soybean meal, 48% CP	31.16	26.11	22.25
Restaurant grease	2.60	5.65	5.50
Dicalcium phosphate	1.65	1.80	1.40
Calcium carbonate	0.97	0.92	1.21
Copper sulfate	0.05	0.07	0.05
Mineral premix <sup>3</sup>	0.15	0.15	0.10
Vitamin premix <sup>4</sup>	0.15	0.15	0.125
${ m Antibiotic}^5$	0.125	0.125	0.05
Ethoxyquin	0.03	0.03	0.03
Salt	0.50	0.50	0.50
Calculated composition, %			
Crude protein	19.88	17.62	16.09
Lysine	1.10	0.96	0.85
Threonine	0.78	0.69	0.63
Tryptophan	0.27	0.23	0.21
Methionine + Cystine	0.65	0.59	0.55
Calcium	0.80	0.80	0.70
Phosphorus	0.69	0.69	0.60

<sup>1</sup>Basal diets were supplemented with 0.05% of a *Bacillus*-based direct-fed microbial to provide 2 experimental diets.

<sup>2</sup>Pigs were fed the starter diet from 23 to 36 kg of BW, the grower diet from 36 to 64 kg of BW, and the finisher diet from 64 kg of BW to market weight.

<sup>3</sup>Supplied 84 mg of Ca as calcium carbonate, 165 mg of Fe as ferrous sulfate, 165 mg of Zn as zinc sulfate, 39 mg of Mn as manganese sulfate, 16.5 mg of Cu as copper sulfate, 0.30 mg of I as calcium iodate, and 0.30 mg of Se as sodium selenite per kilogram of complete feed during the starter and grower phases, and 70 mg of Ca as calcium carbonate, 137.5 mg of Fe as ferrous sulfate, 137.5 mg of Zn as zinc sulfate, 32.5 mg of Mn as manganese sulfate, 13.75 mg of Cu as copper sulfate, 0.25 mg of I as calcium iodate, and 0.25 mg of Se as sodium selenite per kilogram of complete feed during the finisher phase

 $^4\mathrm{Supplied}\ 238.5\ \mathrm{mg}$  of Ca as calcium carbonate, 6,613.8 IU of vitamin A, 992.1 IU of vitamin D\_3, 26.46 IU of vitamin E, 0.0265 mg of vitamin B\_{12}, 2.65 mg of vitamin K (menadione), 5.95 mg of riboflavin, 19.84 mg of pantothenic acid, and 33.07 mg of niacin per kilogram of complete feed during the starter and grower phases, and 159 mg of Ca as calcium carbonate, 4,409.2 IU of vitamin A, 661.4 IU of vitamin D\_3, 17.64 IU of vitamin E, 0.0176 mg of vitamin B\_{12}, 1.76 mg of vitamin K (menadione), 3.97 mg of riboflavin, 13.23 mg of pantothenic acid, and 22.05 mg of niacin per kilogram of complete feed during the finisher phase.

<sup>5</sup>Provided 0.11 g of tylosin per kilogram of feed during the starter and grower phases, and 0.04 g of tylosin per kilogram of feed during the finisher phase.

tions according to the NRC (1998). A 3-phase finishing program was implemented in each study, with diet transitions occurring from starter to grower and grower to finisher phases when the mean BW of each block averaged 34 and 68 kg, respectively. Pigs were removed from the study as the mean BW of each pen reached approximately 104 kg. Average pig BW and feed disappearance were determined for each pen at the end of the starter, grower, and finisher phases and ADG, ADFI, and G:F were calculated.

At the completion of the third study, 2 sections of the solid manure buildup (manure mat) in the solid portion of the pen and weighing approximately 100 g were ob-

tained from 2 locations in 24 pens (12 pens/treatment) housing 6 pigs/pen. A small rectangular manure mat sample weighing approximately 4.0 g was cut from each 100-g manure mat section. The samples were placed separately in a beaker with 500 mL of water at room temperature (approximately 25°C) with a magnetic stir bar. The beaker was placed on a magnetic stir plate, and the time required to disperse the solid manure mass with stirring action as evidenced by visual inspection was determined. Values from the 2 manure mat samples from each pen were averaged to derive a mean dispersal time (min/g of manure mat sample) for each pen. After acquiring the manure mat samples, the pens were cleaned by 4 individuals randomly assigned to clean 6 of the 24 pens. Each individual cleaned 3 control and 3 treated pens and was unaware of the treatments assigned to each pen. The pens were scraped with a scraper with a 17.8-cm blade and then were washed with unheated tap water dispersed at 13.8 MPa by a high-pressure washer. The time required to clean each pen was determined by measuring with a stopwatch and summing the total time required to scrape the manure buildup from the pen floor surface and to wash with a high-pressure washer.

Data were analyzed as a randomized complete block design, with pen as the experimental unit and blocks based on initial BW. Analysis of variance was performed using the MIXED procedure (SAS Inst. Inc., Cary, NC), and the effects of experiment, dietary treatment, and the experiment × treatment interaction were evaluated as fixed effects. Block within experiment was a random effect. When a significant interaction was observed, treatment means were separated using the PDIFF option of the LSMEANS statement of PROC MIXED. Because an experiment × treatment interaction was detected in only 1 study, the main effect means pooled across experiments are reported.

## Commercial Swine Facility Evaluation

An additional study was conducted on a large, commercial swine operation over an approximately 16-mo period. All-in, all-out, wean-to-finish sites within the commercial operation were randomly assigned as control or treated sites. Average daily gain, G:F, and death loss percentage were monitored at each site using record-keeping software (PigChamp Inc., Ames, IA). Data were collected at each site as pig groups left the facility for market. Initially, both sites were fed common cornsoybean meal-based diets typical of those fed in commercial swine production for 7 mo, and the data were collected to serve as a baseline period for comparison. The baseline period began in November 2002 and continued through May 2003. After this 7-mo baseline period, the treated sites were administered Bacillus supplementation at 0.05% of the diet, whereas the control sites continued to be fed the same diet devoid of the Bacillus supplement. The treatment period began in June 2003 for both sites and was completed in January

2004 at the control sites, and in December 2003 at the Bacillus-treated sites.

A total of 16 sites were represented in the control baseline period, during which 34 observations were derived from 33 barns (2 separate groups of pigs closed out of 1 barn in the 7-mo period). Thirteen sites were represented in the control treatment period, during which 34 observations were derived from 29 barns. A total of 18 sites were represented in the treated baseline period, during which 32 observations were derived from 32 barns. The *Bacillus*-supplemented group was represented by 12 sites during the treatment period, with 23 observations derived from 23 barns.

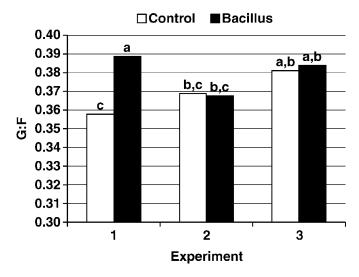
Measurements of each performance variable as each barn was emptied at market weight (considered a settlement of pigs) in the control and treated sites for the 16-mo period were imported into Statistical Process Control software (BaRaN Systems Ltd., Sherwood Park, Alberta, Canada). An analysis of the mean and SD for each performance measurement was conducted separately for the baseline and the treatment periods. The mean and SD were used to set a lower control limit (LCL) and an upper control limit (UCL) based on 2 SD from the mean for control and treated sites during the baseline and treatment periods. Data were plotted on the control charts to evaluate the location of the data points in relation to the mean, LCL, and UCL. Symbols used to plot the data points within the control charts were defined as follows: (•) the data point lies within  $\pm 2$  SD from the mean, or ( $\bigcirc$ ) the data point lies outside ± 2 SD from the mean. Data points that were outside the LCL and UCL were interpreted as violating the expected output trends and were stated to be "out of statistical control", meaning that an unexpected outcome that deviated from the expected normal range of outputs had occurred.

#### **RESULTS**

## University Research Experiment

*Growth Performance*. Supplementation with *Bacillus* in Exp. 1 increased (P = 0.002) G:F in the overall trial (starter to market weight) compared with control pigs, whereas there was no difference observed between Bacillus-supplemented and control pigs in Exp. 2 and Exp. 3 (experiment  $\times$  treatment interaction, P = 0.03; Figure 1). Due to the presence of only 1 significant experiment × treatment interaction, data from the 3 experiments were combined to evaluate the effect of Bacillus supplementation across the 3 studies. In the combined analysis of the 3 experiments, ADG and ADFI did not differ  $(P \ge 0.16)$  between the 2 dietary treatments (Table 2). However, G:F increased (P = 0.04)when pigs were fed diets containing Bacillus compared with control diets during the finisher phase and in the overall growing-finishing period.

Manure Dispersibility. Manure mat samples obtained from pens housing pigs fed Bacillus dispersed



**Figure 1.** Gain:feed of pigs fed a control or *Bacillus*-supplemented diet during the growing-finishing period of Exp. 1, 2, and 3 of the university research study. Values represent the mean of each treatment for each of the 3 experiments, with a total of 8 pens/treatment in Exp. 1 and 2, and 12 pens/treatment in Exp. 3 (experiment × treatment interaction, P = 0.03). <sup>a-c</sup>Means lacking a common letter differ (P < 0.05; SE = 0.006 for Exp. 1, 2, and 3).

**Table 2.** Main effect responses of pigs fed control diets or *Bacillus*-supplemented diets throughout the starter (23 to 36 kg of BW), grower (36 to 64 kg of BW), and finisher (64 kg to market weight) phases during the university research experiment<sup>1</sup>

Treat	$Treatment^2$		P-value for
Control	Bacillus	SE	treatment effet
0.569	0.575	0.012	0.69
1.373	1.356	0.030	0.66
0.415	0.425	0.007	0.29
0.840	0.839	0.016	0.94
2.154	2.121	0.035	0.31
0.391	0.395	0.004	0.43
0.957	0.976	0.015	0.28
2.895	2.820	0.051	0.16
0.331	0.350	0.006	0.04
0.840	0.845	0.011	0.54
2.279	2.228	0.038	0.12
0.369	0.380	0.003	0.03
	0.569 1.373 0.415 0.840 2.154 0.391 0.957 2.895 0.331 0.840 2.279	Control         Bacillus           0.569         0.575           1.373         1.356           0.415         0.425           0.840         0.839           2.154         2.121           0.391         0.395           0.957         0.976           2.895         2.820           0.331         0.350           0.840         0.845           2.279         2.228	Control         Bacillus         SE           0.569         0.575         0.012           1.373         1.356         0.030           0.415         0.425         0.007           0.840         0.839         0.016           2.154         2.121         0.035           0.391         0.395         0.004           0.957         0.976         0.015           2.895         2.820         0.051           0.331         0.350         0.006           0.840         0.845         0.011           2.279         2.228         0.038

<sup>&</sup>lt;sup>1</sup>The 3 university research experiments were combined for analysis and means representing the combined data are displayed. Values are the means of 28 pens representing each of the 2 dietary treatments.

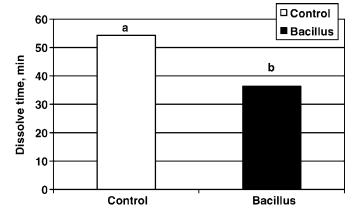


Figure 2. Time required to dissolve manure mat samples collected from a total of 24 pens (12 pens/treatment) housing pigs fed a control or Bacillus-supplemented diet during Exp. 3 conducted at a university research facility. Two sections of the solid manure build-up (manure mat) in the solid portion of the pen weighing approximately 100 g were obtained from 2 pen locations. A small rectangular manure mat sample weighing approximately 4.0 g was cut from each 100-g manure mat section. Samples were placed in 500 mL of water at room temperature with a magnetic stir bar and the time required to disperse the solid manure mass with stirring action as evidenced by visual inspection was determined. Values from the 2 manure mat samples from each pen were averaged to derive a mean dispersal time (min/g of manure mat sample) for each pen. a,b Means lacking a common letter differ (P =0.03; SE = 5.64).

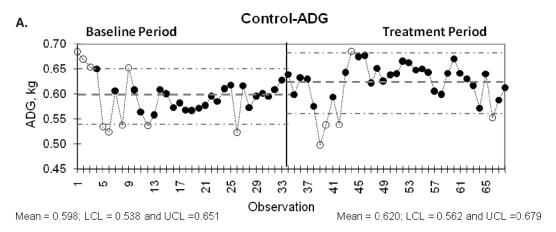
in less (P=0.03) time than those obtained from pens fed the control diet, representing a 33% reduction in the time required to dissolve the manure mat with Bacillus supplementation (Figure 2). Although the time required to wash pens housing control or Bacillus-treated pigs did not differ (8.7 and  $8.5\pm0.9$  min, respectively; P=0.87), the quicker dispersal time of the manure mat from Bacillus-treated pens was supported by numerically less total time required to clean (scrape and wash) pens housing Bacillus-supplemented pigs (23.0 vs. 21.0  $\pm$  2.2 min; P=0.52).

# Commercial Swine Facility Evaluation

Evaluation of *Bacillus* supplementation in a commercial swine facility using SPC is illustrated in Figures 3, 4, and 5, in which the mean, UCL, and LCL are displayed for the baseline period and the treatment period. The mean ADG observed for each settlement of pigs from control sites during the baseline period was slightly greater than the mean ADG during the baseline period at sites treated with *Bacillus* (Figure 3). However, the mean ADG during the treatment period for both the control and treated sites increased to 0.620 and 0.621 kg, respectively, resulting in an improvement of 4.4% at control sites and 7.6% at sites supplemented

<sup>&</sup>lt;sup>2</sup>Dietary treatments were comprised of a control basal diet and the control diet supplemented with 0.05% of a Bacillus-based DFM that provided  $1.47 \times 10^8$  cfu of Bacillus organisms per gram of supplement.

<sup>&</sup>lt;sup>3</sup>Combined data representing the overall effect from the beginning of the trial (starter phase) to the completion of the finisher phase (market weight).



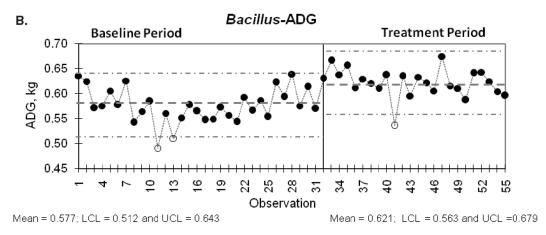


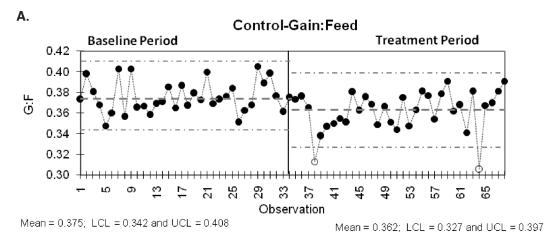
Figure 3. Control charts from statistical process control analysis of ADG of pigs fed (A) a control or (B) *Bacillus*-supplemented diet within a commercial swine production facility. Data are plotted on the control charts to evaluate the location of data points in relation to the mean, lower control limit (LCL), and upper control limit (UCL). Symbols used to plot the data points within the control charts were defined as follows: (●) data point lies within ±2 SD from the mean, or (○) data point lies outside ±2 SD from the mean. Data are plotted to illustrate how each observation compares to the expected average (dashed line) of the operation and the upper and lower control limits (UCL and LCL; dot-dash line). The baseline period in both graphs represents data collected from sites randomly selected for each treatment (34 observations from control sites and 32 observations from treated sites) for 7 mo before the administration of dietary treatments, whereas the treatment period represents observations after treatments were administered.

with *Bacillus*. Additionally, of the 5 settlements from control sites that fell out of the UCL and LCL during the treatment period, 4 were below the LCL. Only 1 settlement treated with *Bacillus* was below the LCL.

Administration of *Bacillus* to the treated sites had minimal effect on G:F (Figure 4). The mean G:F ratio was lower during the baseline period for sites assigned to receive *Bacillus* supplementation compared with control sites and although no control settlement observation for G:F placed out of the UCL or LCL during the baseline period, 4 G:F measurements from sites assigned to *Bacillus* treatment were below the LCL during this period. During the treatment period, the mean G:F ratio for control sites decreased slightly from the baseline period, and 2 settlements were observed to be below the LCL. The mean G:F ratio during the treatment period at *Bacillus*-treated sites was similar to that observed during the baseline period. Additionally,

during the treatment period 1 settlement from *Bacillus*-supplemented sites fell above the UCL and 3 fell below the LCL during the treatment period.

Mean death loss percentage at control sites was lower compared with sites assigned to *Bacillus* supplementation during the baseline period (Figure 5). The mean death loss of control sites during the treatment period increased to 6.8% whereas a decrease in death loss to 5.1% was observed for *Bacillus*-treated sites during the treatment period. This resulted in a 17.5% increase in death loss percentage at control sites and a 40.9% decrease at treated sites compared with the baseline period. Three control settlement measurements fell above the UCL during the baseline period compared with 4 settlements from *Bacillus*-supplemented sites during the same period. During the treatment period, 3 control settlement measurements for death loss per-



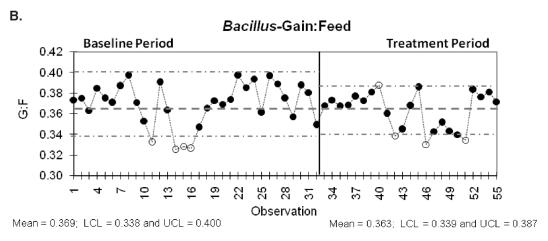


Figure 4. Control charts from statistical process control analysis of gain:feed of pigs fed (A) a control or (B) *Bacillus* supplemented diet within a commercial swine production facility. Data are plotted on the control charts to evaluate the location of data points in relation to the mean, lower control limit (LCL), and upper control limit (UCL). Symbols used to plot data points within control charts were defined as follows: ( $\bullet$ ) data point lies within  $\pm 2$  SD from the mean, or ( $\bigcirc$ ) data point lies outside  $\pm 2$  SD from the mean. Data are plotted to illustrate how each observation compares to the expected average (dashed line) of the operation and the upper and lower control limits (UCL and LCL; dot-dash line). The baseline period in both graphs represents data collected from sites randomly selected for each treatment (34 observations from control sites and 32 observations from treated sites) for 7 months before the administration of dietary treatments, whereas the treatment period represents observations after treatments were administered.

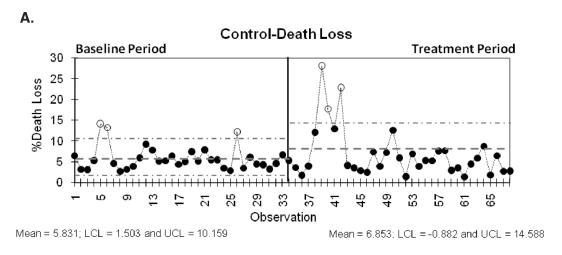
centage were above the UCL compared with only one of the settlements from treated sites.

## **DISCUSSION**

There are several remedies available to aid in handling, processing, and management of swine waste that are mostly targeted toward alleviating odorous compounds and decreasing nitrogen loss through the volatilization of ammonia. These approaches include dietary alterations to reduce or alter N excretion in urine and feces, gastrointestinal microbial manipulation to decrease excretion of malodorous compounds, feed additives to alter manure pH and ammonia losses, and pit additives to enhance manure break-down (Sutton et al., 1999; Zhu, 2000; Smith et al., 2004). Most of these approaches fail to address the primary problem of inadequate microbial decomposition of manure, and micro-

bial pit additives are difficult to administer such that the effective microorganisms are consistently distributed within manure storage facilities. Several studies in the scientific literature document survival of *Bacillus* spores through the digestive process, germination of *Bacillus* spores within the digestive tract, and excretion of *Bacillus* spores through fecal matter (Hoa et al., 2001; Casula and Cutting, 2002; Nicholson, 2002). This study demonstrates that administration of a direct-fed microbial additive to swine diets is an effective means of distributing microorganisms specifically selected to promote solids degradation in manure pits.

Manure build-up typical of partially slatted pens was evident during each of the 3 experiments; therefore, two 100-g manure mat sections were easily obtained from each pen in Exp. 3. The high degree of variation observed in the cleaning time measurements due to 4 individuals cleaning the pens and the factor of fatigue



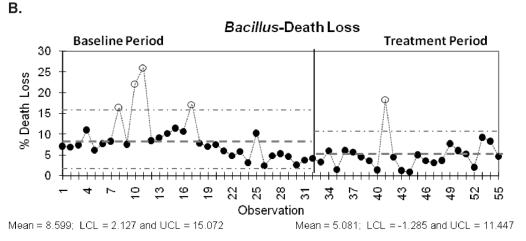


Figure 5. Control charts from statistical process control analysis of death loss percentage of pigs fed (A) a control or (B) *Bacillus*-supplemented diet within a commercial swine production facility. Data are plotted on the control charts to evaluate the location of data points in relation to the mean, lower control limit (LCL), and upper control limit (UCL). Symbols used to plot data points within control charts were defined as follows: (●) data point lies within ±2 SD from the mean, or (○) data point lies outside ±2 SD from the mean. Data are plotted to illustrate how each observation compares to the expected average (dashed line) of the operation and the upper and lower control limits (UCL and LCL; dot-dash line). The baseline period in both graphs represents data collected from sites randomly selected for each treatment (34 observations from control sites and 32 observations from treated sites) for 7 months before the administration of dietary treatments, whereas the treatment period represents observations after treatments were administered.

throughout the process resulted in the inability of this model to detect a statistically significant difference in the time required to manually clean control and treated pens. The assay to measure the time to disperse manure mat samples was conducted to simulate the pen cleaning process and decrease the variation associated with multiple washers. Less time required to disperse solid manure samples obtained from pens housing pigs supplemented with a Bacillus-based DFM indicates the microorganisms were effective at breaking up manure solids and demonstrates its effectiveness as a manure pit additive. The Bacillus strains present in the DFM were selected for their protease, amylase, and cellulase activities, and these enzymatic activities are a likely explanation for why manure mats from pens treated with the DFM dispersed faster than those from control pens. However, because the nutrient composition of manure mat samples was not measured in this study, how supplementation with the *Bacillus*-based DFM decreased the time required to disperse the manure samples remains inconclusive.

Inclusion of tylosin in the growing-finishing diets had no obvious effect on the ability of the DFM to improve manure characteristics as evidenced by the decreased time required to disperse manure mat samples. Although *Bacillus* organisms exhibit a range of susceptibilities to various antibiotics, in vitro screening of the *Bacillus* strains included in the DFM have been conducted in our laboratory and indicate strains in the DFM are not susceptible to some of the antibiotics commonly used in swine diets, including penicillin and lincomycin (unpublished data). Tylosin was not included

in the in vitro screening procedure; however, the G:F response observed in pigs supplemented with the DFM suggests the *Bacillus* strains present in the DFM remain active in the presence of tylosin.

Improvement in G:F in the latter part of the growingfinishing period resulting from Bacillus supplementation in the university research study may have been due to several functional characteristics of Bacillus microorganisms. Bacillus species have been identified as potent producers of extracellular degrading enzymes, including amylases, cellulases, lipases, and proteases (Ferrari and Schmidt, 1993). Administration of Bacillus organisms in swine feed may provide a source of these enzymes to the pig, aiding in nutrient digestion and utilization of feed and thereby improving growth efficiency. In a previous study, the Bacillus-based directfed microbial evaluated in this study resulted in a decrease in ammonia emissions from simulated anaerobic manure pits (Cromwell et al., 1999) and may better explain the efficiency improvement later in the finishing phase because the microorganisms would have had longer to alter manure composition to favor less ammonia production. Another possibility is that the *Bacillus* organisms may impact pig health through beneficial immune modulation. The role of Bacillus organisms as probiotics for human and animal products has been extensively reviewed, and describes how the production of antimicrobial compounds and alterations in immune cell populations and functions by *Bacillus* species may protect from pathogenic challenges (Hong et al., 2005). More specifically, earlier studies with neonatal and weaned piglets have reported that supplementation with B. subtilis and B. lichenformis decreased the incidence of diarrhea (Maruta et al., 1996; Kyriakis et al., 1999). However, the mechanism by which the Bacillusbased DFM evaluated in this study improved efficiency of growth cannot be determined from these data.

One of the major challenges of implementing new technologies into swine production systems is measuring the benefits of these technologies in commercial field conditions. Traditional statistical methods are at a disadvantage in detecting significant differences between treatments due to the difficulty in obtaining production measurements and the many sources of variation that cannot be defined in the statistical model. Statistical process control is a technique commonly used in manufacturing industries to monitor variation of outputs from the manufacturing process (Grigg, 1998). Many sources of variation that are inherent to the manufacturing process result in a stable, repeatable distribution over time, and when outputs fall within this expected distribution the process is said to be "in statistical control". Special sources of variation will cause the process distribution to change, resulting in unpredictability of the final output and a change in the process distribution. These changes in the distribution can be beneficial or detrimental and should be evaluated such that beneficial inputs can be made a part of the process and detrimental inputs can be identified quickly and eliminated.

Statistical process control provides a means to monitor an agricultural system, much like manufacturing industries monitor their production processes, to detect changes in response variables due to the implementation of new technologies such as the direct-fed microbial feed additive evaluated in this study. Because labor, facilities, and biosecurity limit the number and types of measurements that can be obtained in a large commercial swine facility, the evaluation of the Bacillusbased direct-fed microbial under field conditions was limited to data that could be gleaned from the Pig-Champ software used to evaluate production data within the commercial facility and included only ADG, G:F, and death loss percentage over the growing-finishing period. Improvements in these performance measurements should manifest in SPC analysis as shifts in the expected mean of the treated sites from the baseline period to the treatment period. Additionally, points within the UCL and LCL indicate that measurements from an analyzed site are within an expected range based on the variation within the system. Deviations of measurements outside of this expected range suggest some input has shifted the measurements away from the mean and range around the mean in which measurements would be expected to fall. Inputs which result in measurements outside of the UCL and LCL could be unintentional phenomenon such as a disease outbreak or an intended change such as an alteration in dietary ingredients or formulation. Control sites provide a reference to compare alterations in the expected mean from the baseline period before supplementation with the DFM to the treatment period after the administration of treatment.

Statistical process control analysis conducted in this evaluation detected a decrease in the expected mean for death loss percentage when treatment with the Bacillus-based DFM was implemented. However, the G:F response observed in the controlled university experiment was not detected in the evaluation conducted at a commercial production facility. This may be explained by the experiment × treatment interaction observed for the G:F response in the university study, in which a significant response was observed only in the first experiment with a G:F response in the control group less than 0.36. Like the other 2 experiments, G:F in the commercial facility was greater than 0.36 and greater than the threshold in which increases in performance would be expected. Although the improved ADG detected from the direct-fed microbial treatment in the commercial facility was not detected in the university study, limited challenge conditions in a controlled research facility may have allowed pigs to reach ADG responses closer to their potential compared with conditions in the commercial swine facility in this evaluation.

Data points falling outside of the UCL and LCL were observed during the baseline and treatment time points for both control and treated sites. When all data points

fall within the UCL and LCL, the site would be considered to be "in control", meaning each measurement is falling within a predictable, controlled variation caused by many sources within a swine production facility (Neave and Wheeler, 1996). Therefore, interpreting aberrant data points is difficult, due to the difficulty in attributing a cause as to why data did not fall within the expected range around the mean; however, data points outside the UCL and LCL do indicate that some assignable cause became a source of uncontrolled variation within the site.

Statistical process control has been implemented in the agricultural industry to monitor food safety (Murphy et al., 2005) and animal health (Cowen et al., 1994; Niza-Ribeiro et al., 2004; Baum et al., 2005) and to monitor somatic cell counts in the dairy industry (Niza-Ribeiro et al., 2004; Lucas et al., 2005). Whereas SPC methods have been utilized to effectively monitor response measurements and their variation over time, pinpoint specific "out of control" observations, and discover and remedy these aberrant observations (Neave and Wheeler, 1996), this study used SPC to evaluate whether implementation of a specific technology would provide a long-term benefit for a commercial swine production facility. Whereas the effectiveness of a technology must be verified in controlled research studies, this evaluation demonstrates that SPC can be used to measure the outcome of implementing new technologies in a commercial swine production facility. One caveat to consider when evaluating new technologies in a swine production facility using SPC is the potential effect of seasonality on the response measurements. Whereas this study used 7 mo before implementation of the DFM, it may be more prudent to use historical data that coincides with the same months in which the postimplementation period is being evaluated.

Supplementation with a specifically selected *Bacillus*-based DFM improved feed efficiency and decreased the time required to disperse a swine manure mat sample in a controlled university research study. Furthermore, when evaluated in a commercial swine production facility using statistical process control analysis, the DFM improved gain and mortality of pigs during the growing-finishing period. These data suggest this DFM, composed of *Bacillus* organisms specifically selected to aid in the manure decomposition process, improves pen cleaning in growing/finishing facilities and growth performance of growing-finishing pigs.

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