Title: Evaluating effects of mitigation strategies on pig welfare and future productivity after transport during different seasons – **NPB #16-065**

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Industry Summary:

As consumer concern for swine welfare, husbandry methods, and antibiotic use in animal agriculture increase, so will the need for U.S. pork producers to demonstrate that current production practices not only increase animal performance, but also improve the overall health and welfare of animals. In modern swine production, newly weaned pigs are often subjected to multiple stressors including weaning stress, transport stress, and thermal stress, and these have the potential to increase the incidence of animal disease, morbidity, and mortality, especially when they occur concomitantly. In order to promote stress recovery, improve animal welfare, and prevent the onset of disease, producers often administer dietary antibiotics for 14 to 42 days after pigs enter wean-to-finish facilities. However, due to increased consumer concern regarding the use of antibiotics in animal production, and legislative action promoting antibiotic free diets, it has become increasingly important to develop antibiotic alternatives that can help pigs recover from stressful events as effectively as dietary antibiotics. It was determined that replacing dietary antibiotics with 0.20% L-glutamine improved the productivity of weaned pigs at a similar level as dietary antibiotics throughout the nursery phase. However, the effects of dietary antibiotics and L-glutamine provided d 0-14 post-weaning on pig productivity were diminished as pigs entered the grow-finish phase. In addition, aggressive behavior tended to be reduced overall in L-glutamine fed pigs compared to antibiotic fed pigs, but no differences were observed between antibiotic and L-glutamine treatments versus non-antibiotic pigs. No carcass characteristics were altered by dietary treatments. Pigs weaned in the spring had greater growth performance compared to pigs weaned in the summer resulting in heavier carcass weights and greater loin depth for spring weaned pigs. In conclusion, 0.20% L-glutamine supplementation improved pig health and productivity after weaning and transport similarly to antibiotics; however, the positive growth effects of dietary antibiotics and L-glutamine provided the first 14 days post-weaning were diminished during the grow-finish phase.

Keywords: antibiotics, L-glutamine, pigs, season, transport, weaning

Scientific Abstract:

Antibiotic use has been limited in United States swine production. Therefore, the objective was to determine whether supplementing L-glutamine at cost-effective levels can replace dietary antibiotics to improve piglet welfare and productivity following weaning and transport during different seasons. Based on previous research, we hypothesized that withholding dietary antibiotics would negatively affect pigs while diet supplementation with 0.20% L-glutamine (GLN) would have similar effects on pig performance and health as antibiotics. Mixed sex piglets [N=480; 5.6±0.1kg BW] were weaned (18.4±0.2d of age), and transported for 12 h during summer 2016 and spring 2017. Pigs were blocked by BW and allotted to 1 of 3 dietary treatments [n=10 pens/dietary treatment/season (8 pigs/pen)]; antibiotics [A; chlortetracycline (441ppm) + tiamulin (38.6ppm)], no antibiotics (NA), or GLN fed for 14 d. On d15-34, pigs were provided a common antibiotic free diet. Data were analyzed as a 2×3 factorial of season and diets using PROC MIXED in SAS 9.4. Day 14 BW and d0-14 ADG was greater (*P*=0.01) for A (5.6 and 18.5%, respectively) and GLN pigs (3.8 and 11.4%, respectively) compared to NA pigs, with no differences between A and GLN pigs. Day 0-14 ADFI increased for A (*P*<0.04; 9.3%) compared to NA pigs; however, no differences were detected when comparing GLN to A and NA pigs. Once dietary treatments ceased, no differences (*P*>0.05) in productivity between dietary treatments were detected. Aggressive behavior tended to be reduced overall (*P* = 0.09; 26.4%) in GLN compared to A pigs, but no differences were observed between A and GLN versus NA pigs. Huddling, active, and eating/drinking behaviors were increased overall (*P* < 0.02; 179, 37, and 29%, respectively) in the spring compared to the summer season. When HCW was used as a covariate, loin depth and lean percentage was increased (*P* = 0.01; 4.0% and 1.1%, respectively) during the spring compared to the summer. In conclusion, GLN supplementation improved pig performance and health after weaning and transport similarly to A across seasons; however, the positive effects of A and GLN were diminished when dietary treatments ceased.

Introduction:

Weaning is a complex stressor associated with social, environmental and metabolic stress in pigs (Lallés et al., 2004). In newly weaned pigs, stress is induced by separation from the sow, relocation and mixing piglet groups, and a radical change in diet that often reduces or eliminates feed intake in the first 48 hours post-weaning (Brooks et al., 2001). As a result, piglets undergo a variety of physiological and metabolic changes that can negatively impact welfare. Changes may result from elevated blood cortisol levels (Moeser et al., 2007; Van der Meulen, et al., 2010), compromised feed intake (Maenz et al., 1994), altered intestinal morphology (Lallés et al., 2004), and dehydration due to the switch from an all liquid (milk) to a solid diet (Berry and Lewis, 2001). Unfortunately, in commercial production systems, weaning stress may be compounded by transport stress, which can induce significant weight loss with only 4 h of travel time (Hicks et al., 1998), and ambient temperature likely plays a critical role in determining total stress load incurred by piglets (Lambooy, 1988). Therefore, it is imperative that effective recovery strategies are developed to improve the welfare and productivity of pigs following these stressful events.

Historically, swine producers used dietary antibiotics to help newly weaned pigs overcome the stress of weaning and associated stressors (Chiba, 2010). However, due to increased consumer concern regarding the use of antibiotics in animal production, and legislative action promoting antibiotic free diets, it has become increasingly important to develop alternatives that can help pigs recover from stressful events as effectively as dietary antibiotics. Previous research determined that inclusion of 0.20% L-glutamine (Ajinomoto North America, Inc., Raleigh, NC) in the diets of newly weaned and transported pigs could improve growth rate and well-being more effectively than dietary antibiotics ([chlortetracycline (Aureomycin, Zoetis, Parsippany, NJ) + tiamulin (Denagard, Elanco Animal Health, Greenfield, IN)]; Johnson and Lay, 2017). However, this study was conducted under controlled conditions utilizing simulated transport and individual housing. Therefore, study objectives were to evaluate the impact of replacing dietary antibiotics with 0.20% L-glutamine on swine welfare, growth performance, health status, and carcass characteristics of pigs in a production environment following weaning and transport during different seasons (summer or spring). We hypothesized that withholding dietary antibiotics would negatively impact the overall well-being of piglets, and that diet supplementation with 0.20% L-glutamine would have a similar effect on piglet health and productivity as dietary antibiotics in a production environment.

Objectives:

1.) To evaluate the impact of transport and heat stress following weaning on swine welfare and future productivity when no therapeutic antibiotics are administered in a production environment.

2.) To validate our initial findings in a controlled environment that L-Glutamine, a nutraceutical, will improve piglet welfare more effectively than post-transport therapeutic antibiotic administration and improve future productivity when piglets are exposed to the additive effects of weaning stress, transport stress and heat stress while in a production environment.

Materials & Methods:

General

All procedures involving animal use were approved by the Purdue University Animal Care and Use Committee (protocol #1603001385), and animal care and use standards were based upon the *Guide for the Care and Use of Agricultural Animals in Research and Teaching* (Federation of Animal Science Societies, 2010). Mixed sex crossbred pigs [N = 480; 5.62 ± 0.06 kg initial BW; Duroc x (Landrace x Yorkshire)] were weaned and transported at 18.4 ± 0.2 d of age in central Indiana during the summer of 2016 and the spring of 2017. One day prior to weaning and transport, all pigs were individually weighed, blocked by body weight, and randomly allotted to pens, and pens of pigs were allotted to 1 of 3 dietary treatments with 10 pens per dietary treatment per season of weaning and transport. Pens were blocked by average initial body weight. Each pen, initially, contained 8 pigs. Dietary treatments were antibiotics [**A**; chlortetracycline (441 ppm) + tiamulin (38.6 ppm)], no antibiotics (**NA**), or 0.20% L-glutamine (**GLN**). The treatments were arranged in a 2 × 3 factorial with main effects of season **(summer or spring**) and diet (**A**, **GLN**, **NA**).

On the day of weaning and transport, selected pigs were removed from sows and herded up a ramp into a gooseneck livestock trailer (2.35 × 7.32 m; Wilson Trailer Company, Sioux City, IA) providing 0.07 m^2 per pig and within the range of 0.060-0.084 m^2 per pig required for 4.54 to 9.07 kg pigs, respectively (Federation of Animal Science Societies, 2010). The loading ramp to the trailer was 2.13 m in length providing an 11.0^0 incline, less than the recommended maximum of 20.0^0 (National Pork Board, 2015). Two data loggers (Hobo®; data logger temperature/RH; Onset®; Bourne, MA) were evenly spaced within the trailer to measure ambient temperature (**T_A**) and relative humidity (**RH**) in 5 min intervals. During transport, the trailer T_A during the summer season was 29.4 ± 0.2°C and during the spring was 11.0 ± 0.2°C with trailer RH being 64.3 ± 0.8% and 63.1 ± 0.9%, respectively. Trailers were bedded with wood shavings and ventilation openings were adjusted based on the T_A (National Pork Board, 2015).

Piglets were transported as a group in the trailer for approximately 12 h and 819 km without feed or water. Total transport time was determined by adding loading time, time spent in the trailer, unloading time, and the time it takes to be sorted into their respective pens in the nursery facility. The average time to wean and load the trailer was 55 m. The drivers were the same and followed the identical route for the summer and spring season. Attention was given when developing the transport route that approximately 50 % two-lane roads and 50 % four-lane roads were utilized for transport. The route was approximately 273 km in length and was completed three times during the transport phase for each season. The route took, on average 3 h 16 m to complete. The driver was changed and the truck was refueled after each time the 273 km route was completed. At the conclusion of the 12 h transport, piglets were unloaded from the trailer, individually weighed, and placed into pens. The average time to unload the trailer, weigh the pigs, and place into pens was 1 h 10 m.

Nursery Phase

Following transport, pigs were placed in their assigned pens and provided their respective dietary treatments for 14 d in two phases (d 0 to 14 of the nursery phase). Following the dietary treatment period, all pigs were fed common antibiotic free diets from d 14 to the end of the nursery phase (d 34; Table 1). Diets were corn-soybean meal-based diets provided to meet or exceed nutrient requirements (NRC, 2012) in meal form in four phases during the nursery period (Table 1). Pigs were weighed individually and feeders were weighed every 7 d during the nursery period to determine the response criteria of ADG, ADFI, and G:F.

In addition, therapeutic antibiotic administration was recorded for the duration of the trial (weaning to market). The researchers and research farm staff were trained to identify pigs needing therapeutic injectable antibiotic treatment and were blinded to the study treatments. Pigs were treated when exhibiting clinical signs of illness. Treatment dose, product given, date given, pig and pen identification, and reason administered were recorded. Reason for therapeutic administration was then categorized for post-hoc analysis. Categories were: enteric challenge (e.g. scours or loose watery stool), respiratory challenge (e.g. coughing, thumping, or labored breathing), lameness (e.g. carrying a limb or difficulty walking or swollen joints), un-thriftiness (e.g. BW loss, poor gain, loss of body condition, or rough hair coat), and all other treatments (e.g. Streptococcus suis, skin infection, and abscess).

The nursery facility where the initial 34 d of the trial was conducted contained pens (1.22 m × 1.37 m) that provided initially approximately 0.21 m^2 per pig. All pens contain one 5-hole dry self-feeder and a cup waterer to allow for ad libitum access to feed and water. The nursery barn has a shallow pit for manure storage and completely slatted plastic floors. The nursery room operated on mechanical ventilation using a 4-stage digital controller (Airstream TC5-2V25A, Automated Production Systems, Assumption, IL, USA). During d 0 to 14 post-weaning, the nursery room average daily T_A during the summer season was 31.48 ± 1.82°C and during the spring was 30.57 ± 0.68°C. From d 14 to 34 the nursery T_A was 28.70 ± 1.14°C and 25.99 ± 0.84°C for the summer and spring seasons, respectively.

Animal Behavior

Piglets were video-recorded for 14 d immediately following weaning and transport using ceiling mounted cameras (Panasonic WV-CP254H, Matsushita Electric Industrial Co. Ltd., Osaka, Japan) attached to a digital video recorder system (GeoVision VMS Software; GeoVision Inc., Tapei, Taiwan). Video was recorded both during the light and the dark periods (12 h : 12 h). Video files were later analyzed using Observer XT 11.5 behavioral analysis software (Noldus Information Technology B.V., Wageningen, The Netherlands) by four trained individuals that were blind to the treatments and maintained an agreement of 90% or greater. Individual behaviors were determined using an instantaneous scan sampling technique in 10 min intervals on d 2, 4, 8 and 12 post-weaning for 3 periods each day (0800-1000 h, 1100-1300 h, and 1400-1600 h) for sickness and other behaviors. Sickness behavior include huddling and other behaviors included active, resting, aggressive, eating/drinking, and non-visible. Then, percent of observations each pen displayed the behavior was calculated. A definition for each behavior is defined in an ethogram (Table 2). The absolute temperature range measured on each day of behavior analysis was as follows: d 2 for summer and spring (30.30 – 32.70°C and 27.56 – 32.83°C, respectively), d 4 for summer and spring (29.97 – 36.32°C and 30.60 – 33.61°C, respectively), d 8 for summer and spring (29.42 – 35.43°C and 27.31 – 32.36°C, respectively, and d 12 for summer and spring (28.97 – 36.86°C and 26.12 – 31.36°C, respectively).

Grow-Finish Phase

On day 34, all pigs were moved to the grow-finish facility for the remainder of the trial and pen integrity was maintained. Common antibiotic free diets were corn-soybean meal-DDGS-based diets provided in meal form to meet or exceed nutrient requirements (NRC, 2012) in six phases during the grow-finish period (Table 3). Pigs and feeders were weighed every 21 d during the grow-finish period to determine the response criteria of ADG, ADFI, and G:F.

The grow-finish facility contained pens (1.68 m × 4.27 m) that provided approximately 1.19 m^2 per pig. All pens contained one 2-hole dry self-feeder and a nipple waterer to allow for ad libitum access to feed and water. The grow-finish barn had a shallow pit for manure storage and completely slatted concrete floors. The barn was mechanically ventilated. During d 0 to 62 of the grow-finish phase, the room average daily T_A during the summer season was 22.35 ± 1.14°C and during the spring was 25.47 ± 2.64°C. From d 62 to 125 the T_A was 19.87 ± 0.83°C and 25.74 ± 2.48°C for the summer and spring seasons, respectively.

Marketing

At the end of the 159 d experiment, pigs from each pen were individually tattooed with pen number and shipped approximately 48 km to Indiana Packers Corporation (Delphi, IN). Pigs were slaughtered under commercial conditions with carbon dioxide stunning. Standard carcass criteria of loin and backfat depth, hot carcass weight (**HCW**), fat-free lean index, and yield were collected. Fat depth and loin depth were measured with an optical probe (Fat-O-Meater, SFK Technology A/S, Herlev, Denmark) inserted between the third and fourth rib from the last rib (counting from the posterior of the carcass) and 7 cm from the dorsal midline of the hot carcass. Lean percentage was calculated according to the Indiana Packers Corporation (2015) formula and the fat-free lean percentage was calculated according to Schinckel et al. (2010) procedures.

Statistics

Data were analyzed as a randomized complete block design using the PROC MIXED procedure in SAS 9.4 (SAS Institute INC., Cary, NC), with pen as the experimental unit. Main effects of season and diet and their interactions were tested. The assumptions of normality of error, homogeneity of variance, and linearity were confirmed post-hoc. All treatment rate and behavioral data were log-transformed to meet assumptions of normality; however, all log-transformed data are presented as arithmetic means for ease of interpretation. All non-transformed data is presented as LS means. For repeated analyses for growth performance, each pen's respective parameter was analyzed using repeated measures with an auto-regressive covariance structure with week as the repeated effect when required. Statistical significance was defined as $P \leq 0.05$ and a tendency was defined as $0.05 < P \leq 0.10$.

Results:

Growth Performance

Nursery Phase

When comparing the main effect of dietary treatment, ADG was greater overall (P = 0.01; 14.9%) from d 0 to 14 of the nursery period in A and GLN pigs compared to NA pigs, but no ADG differences were detected between A and GLN pigs (Table 4). Overall, from d 0 to 34 of the nursery period, ADG was increased (P = 0.01; 7.9%) in A compared to NA pigs, but no differences were detected between A and NA versus GLN pigs (Table 4). An increase in ADFI (P = 0.04) was detected from d 0 to 14 of the nursery phase for A compared to NA pigs, but no differences were observed between A and NA versus GLN pigs (Table 4). Average daily feed intake tended to be greater (P = 0.09) from d 0 to 34 of the nursery period in A compared to NA pigs, but no differences were observed between A and NA versus GLN pigs (Table 4). Feed efficiency (G:F) was greater overall (P = 0.01; 7.7%) from d 0 to 14 of the nursery phase for A compared to NA and GLN pigs, but no differences were observed between NA and GLN pigs (Table 4). From d 0 to 34 of the nursery phase, G:F was increased (P = 0.01; 4.3%) in A compared to NA pigs, but no differences were observed for A and NA pigs compared to GLN pigs (Table 4). Day 14 BW was greater overall (P = 0.01) for A (8.65 ± 0.52 kg) and GLN (8.50 ± 0.52 kg) pigs compared to NA (8.19 ± 0.52 kg) pigs; however, no differences were detected between A and GLN pigs (Table 4). Final BW was increased (P = 0.04) for A (17.78 ± 0.74 kg) compared to NA (16.96 ± 0.74 kg) pigs, but no differences were detected between A and NA versus GLN (17.49 ± 0.74 kg) pigs (Table 4). No other dietary treatment growth performance differences (P > 0.05) were detected during the nursery phase.

A main effect of season was observed where ADFI tended to be reduced (P = 0.08; 5.1%) during the spring compared to the summer from d 0 to 14 of the nursery phase (Table 4). From d 14 to 34 of the nursery phase, ADG tended to be reduced (P = 0.09) and G:F was reduced (P = 0.01) during the summer compared to the spring (3.7 and 7.4%, respectively; Table 4). Overall, from d 0 to 34 of the nursery period, G:F was reduced (P = 0.04; 4.1%) during the summer compared to the spring (Table 4). No other seasonal effects were observed during the nursery period (P > 0.05).

A diet x season interaction was detected (P = 0.04) from d 14 to 34 of the nursery phase where G:F was greater in the spring in NA (0.69 ± 0.01) and GLN (0.68 ± 0.01) pigs compared to NA pigs (0.61 ± 0.01) during the summer. However, no differences were observed between A pigs (0.66 ± 0.01) during the spring and A (0.64 ± 0.01) and GLN (0.63 ± 0.01) pigs during the summer (data not presented). No other diet x season differences were detected (P < 0.05).

Grow-Finish Phase

No dietary treatment differences were observed (P > 0.17) during the grow-finish period (Table 5). From d 0 to 62 of the grow-finish phase, G:F was reduced (P = 0.01; 4.3%) during the summer compared to the spring (Table 5). Average daily gain, ADFI, and G:F were reduced (P = 0.01; 14.6, 4.4, and 12.1%, respectively) during the summer compared to the spring from d 62 to 125 of the grow-finish phase (Table 5). Overall, from d 0 to 125 of the grow-finish period, ADG and G:F were reduced (P = 0.01; 9.2 and 5.1%, respectively) in the summer compared to the spring (Table 5). Final BW at the end of the grow-finish period was reduced (P = 0.01; 9.82 kg) in the summer compared to the spring (Table 5). No other growth performance differences were observed (P > 0.05) during the grow-finish period with any comparison (Table 5).

Behavior

Aggressive behavior tended to be reduced overall (P = 0.09; 26.4%) in GLN compared to A pigs, but no differences were observed between A and GLN versus NA pigs (Table 6). No other diet differences were observed for behavior (P > 0.05) with any comparison (Table 6).

Huddling, active, and eating/drinking behaviors were increased overall (*P* < 0.02; 179, 37, and 29%, respectively) in the spring compared to the summer (Table 6; Fig. 1A, 1B, 1E). Non-visible behavior was greater (*P* < 0.04; 121%) in the summer compared to the spring (Table 6; Fig. 1F). No other season differences were observed for behavior (*P* > 0.05) with any comparison (Table 6; Fig. 1).

Huddling behavior was greater overall (*P* < 0.01) on d 2 and d 4 (15.58 ± 1.92% and 16.86 ± 2.41%, respectively) compared to d 8 and d 12 (6.07 ± 1.34% and 4.70 ± 1.00%, respectively; Fig. 1A). Active behavior was greater overall (*P* < 0.01) on d 2 (13.44 ± 0.82%) compared to d 4, d 8, and d 12 (8.68 ± 0.54%, 10.54 ± 0.67%, and 10.77 ± 0.49%, respectively; Fig. 1B). In addition, active behavior was greater overall (*P* < 0.01) on d 8 and d 12 compared to d 4 (Fig. 1B). Resting behavior was greater overall (*P* < 0.01) on d 4, d 8, and d 12 (78.32 ± 1.55%, 75.92 ± 1.58%, and 76.67 ± 1.18%, respectively) compared to d 2 (70.19 ± 1.84%; Fig. 1C). Aggressive behavior was greater overall (*P* < 0.01) on d 2 (2.81 ± 0.35%) compared to d 4, d 8, and d 12 (1.33 ± 0.26%, 1.16 ± 0.23%, and 0.76 ± 0.18%, respectively; Fig. 1D). In addition, aggressive behavior was greater overall (*P* < 0.01) on d 4 compared to d 12 but no differences were observed on d 4 and d 12 versus d 8 (Fig. 1D). Eating/Drinking behavior was greater overall (*P* = 0.01) on d 8 and d 12 (10.58 ± 0.59% and 10.33 ± 0.63%, respectively) compared to d 2 and d 4 (9.87 ± 1.02% and 9.30 ± 0.55%, respectively; Fig. 1E). No other day differences were observed for behavior (*P* > 0.05) with any comparison (Fig. 1).

Active behavior was greater (*P* < 0.01; 47.1, 64.9, and 65.2%, respectively) on d 2, d 4 , and d 8 during the spring compared to the summer, but was not different on d 12 (Fig. 1B). Resting behavior was increased (*P* < 0.01; 14.7%) on d 2 during the summer compared to the spring (Fig. 1C); however, on d 12, resting behavior was greater (6.8%) during the spring compared to the summer (Fig. 1C). Aggressive behavior tended to be greater (*P* = 0.07; 85.2%) on d 8 during the spring compared to the summer (Fig. 1D). Eating/drinking behavior was greater (*P* < 0.01) on d 2 (64.8%) and d 4 (92.5%) during the spring compared to the summer, but no differences were detected on d 8 and d 12 (Fig. 1E). No other behavioral differences were detected (*P* < 0.05) with any comparison (Table 6; Fig. 1).

Treatment rate

Nursery Phase

A diet x season effect was detected (*P* = 0.04) where pigs treated for lameness from d 14 to 34 was greater in the spring for GLN pigs (2.12 ± 1.00%) compared to all other treatments. However, no differences were observed between A (0.56 ± 1.00%) and NA (0.00 ± 1.00%) pigs during the spring, and A (0.48 ± 1.00%), GLN (0.00 ± 1.00%), and NA (0.00 ± 1.00%) pigs during the summer (Table 7). There were no dietary treatment differences observed (*P* > 0.05) from d 0 to 14 and 0 to 34 (Table 7).

Pigs treated for other reasons were greater (*P* ≤ 0.02) from d 0 to 14 and from 0 to 34 during the spring (1.08 ± 0.46% and 0.53 ± 0.21%, respectively) compared to the summer (0.00 ± 0.46% and 0.00 ± 0.21%, respectively), regardless of dietary treatment (Table 7). No other seasonal effects were observed (*P* > 0.05) for treatment rate (Table 7).

From d 0 to 14, GLN pigs tended (*P* = 0.07) to be treated for enteric challenges more often in the spring (8.19 ± 2.31%) compared to A pigs (3.13 ± 2.31%), and A (3.13 ± 2.31%) and GLN (3.75 ± 2.31%) pigs during the summer (Table 7). No other diet x season differences were detected (*P* < 0.05) during the nursery phase (Table 7).

Grow-Finish Phase

From d 0 to 125 and d 62 to 125, treatment for unthriftiness was reduced (*P* = 0.01) in GLN (0.00 ± 0.54% and 0.00 ± 0.37%, respectively) and NA pigs (0.00 ± 0.54% and 0.31 ±

0.37%, respectively) compared to A pigs (1.11 ± 0.54% and 1.00 ± 0.37%, respectively), but no differences were observed between GLN and NA pigs (Table 8). During d 62 to 125, enteric disease treatments tended ($P < 0.08$) to be reduced by A (0.00 ± 0.93%) treatment and highest for the GLN (1.17 ± 0.93%) treatment with NA (0.34 ± 0.93%) pigs being intermediate (Table 8). No other treatment rate differences for the main effect of dietary treatment were observed ($P >$ 0.05) with any comparison (Table 8).

Pigs treated for lameness were greater ($P < 0.02$) from d 0 to 62, d 62 to 125, and d 0 to 125 during the summer (1.00 ± 0.54, 1.19 ± 0.54, and 1.09 ± 0.32%, respectively) compared to the spring (0.00 ± 0.54, 0.00 ± 0.54, and 0.00 ± 0.32%, respectively), regardless of dietary treatment (Table 8). Treatment for respiratory challenges were greater ($P < 0.01$) from d 0 to 62 and from d 0 to 125 during the summer (666.2 and 138.8%, respectively) compared to the spring (Table 8). Pigs treated for other challenges were greater ($P < 0.02$) during the summer compared to the spring from d 0 to 62 (736.8%) and from d 0 to 125 (888.8%; Table 8). From d 0 to 125, pigs treated for enteric challenges tended to be greater ($P < 0.10$; 250.0%) during the spring compared to the summer (Table 8). No other seasonal effects were observed ($P > 0.05$) for treatment rate (Table 8).

Carcass Characteristics

No dietary treatment effects were observed ($P > 0.60$) on carcass characteristics (Table 9). Hot carcass weight and loin depth were increased ($P < 0.01$; 5.4% and 5.5%, respectively) and carcass yield was reduced ($P < 0.01$; 2.0%) during the spring compared to the summer when HCW was not used as a covariate in the statistical model (Table 9). When HCW was used as a covariate in the statistical analysis, loin depth and lean percentage were increased ($P = 0.01$; 4.0% and 1.1%, respectively) and carcass yield was reduced ($P = 0.01$; 2.3%) during the spring compared to the summer (Table 9). Fat-free lean percentage during the spring tended to be greater ($P = 0.07$; 1.3%) compared to the summer when HCW was included as a covariate (Table 9). No other carcass characteristic differences were observed ($P > 0.05$) with any comparison (Table 9).

Discussion:

The need to wean and transport pigs is necessary to reduce the risk of infectious disease through multi-site production (Harris, 2000). However, the resultant stress response can reduce growth performance and welfare in newly weaned pigs (Chambers and Grandin, 2001; Campbell et al., 2013), especially in the absence of dietary antibiotics (Heo et al., 2013). Despite this, the use of in-feed antibiotics has been reduced in swine production due to consumer preference, legislative action, and concerns about antibiotic resistance (Smith et al., 2010), putting the welfare and productivity of newly weaned and transported pigs at risk and necessitating the development of effective alternatives. Recent work has described improved welfare and productivity in piglets provided GLN compared to A and NA following weaning and simulated transport (Johnson and Lay, 2017). In accordance with the aforementioned study, piglets provided GLN after weaning and transport in the present study had improved growth performance compared to NA pigs during the 14 d dietary treatment period, regardless of season of transport. However, no growth performance differences were detected between GLN and A pigs in our current study. Although reasons for this discrepancy are currently unknown, it may be due to differences in study design since the transport procedure was simulated and piglets were individually housed in the previous study (Johnson and Lay, 2017). While the mechanism(s) of action for improved growth performance has yet to be discerned, GLN can serve as energy source for enterocytes thus reducing jejunal atrophy and intestinal epithelial damage (Wu et al., 1996; Yi et al., 2005). Therefore, it is possible that piglets provided supplemental GLN had improved intestinal barrier function leading to greater pathogen resistance, reduced translocation of bacteria (Peng, 2004) and subsequently an improvement in growth performance (Jiang et al., 2009; Johnson and Lay, 2017). Nevertheless, the advantages observed in early nursery growth performance may suggest that GLN supplementation could serve as an alternative to dietary antibiotics in production systems.

Although growth performance was improved in GLN and A pigs during the dietary treatment period and the advantage was maintained for the overall nursery period, no differences were detected when compared to NA pigs from d 14 to market when all pigs were fed a common antibiotic free diet. However, these results were expected as previous studies have described a loss of growth performance differences once dietary antibiotic treatments (Skinner et al., 2014) or dietary formulation treatments (Dritz et al., 1996) cease. This may be due to pen to pen variability differences that diminished the growth rate advantages as the studies progressed or the performance advantages of feeding dietary treatments are limited only to the period when fed. Therefore, it could be suggested that feeding GLN to pigs for a longer duration could have extended the growth benefits; however, further work would be needed to confirm this hypothesis and any increase in growth performance would need to be balanced with the cost of including GLN in diets.

Weaning and transport is stressful to piglets and may result in behavioral changes including increased aggression and activity that are indicative of distress (Lewis and Berry, 2006; Wamnes et al., 2008). As such, newly weaned and transported piglets in the present study exhibited behavioral signs of distress immediately following transport, which subsequently declined as time progressed following weaning and transport. These behaviors ranged from increased activity, which may be indicative of greater exploratory behavior and stress (Bøe, 1993), to greater huddling behavior that may have been due to greater sub-clinical illness (Hennessy et al., 2001), and an increase in aggressive behavior likely due to fighting and establishing a social hierarchy (Meese and Ewbank, 1973; Blackshaw et al., 1987; Coulson et al, 2012). However, despite the improved growth performance, dietary A and GLN supplementation treatments did not appear to alleviate this post-weaning and transport behavioral stress response relative to NA treated pigs. In addition, aggressive behavior tended to be greater in A compared to GLN pigs, which may be a sign of resource guarding (i.e., feed; Drake et al., 2008) in group-housed pigs. Therefore, potential mechanisms may have been that A pigs spent more time guarding feed as this was the only resource available in the pen or that they felt better and were therefore more capable of doing so. However, because GLN and A pigs had similar ADFI, and NA pigs had reduced ADFI but more aggression, it is still unclear whether the increase in aggressive behavior was due to resource guarding and further research should be performed to determine the cause.

In addition to the impact of weaning and transport as well as dietary treatments on piglet behavior, seasonal effects were also observed. Increased resting behavior was observed during the summer on d 2 post-weaning and transport compared to the spring and this may have been due to greater exhaustion and dehydration during the summer as previously reported (Berry and Lewis, 2001). Furthermore, pigs weaned and transported in the spring exhibited greater huddling behavior compared to those weaned and transported in the summer. Although a specific reason has yet to be elucidated, this response may have been related to T_A and pigs' need for supplemental heat (Hay et al., 2001). This is because the nursery T_A during the summer was at the upper end of the recommended thermoneutral zone and the spring nursery T_A was at the lower end of the recommended thermoneutral zone for nursery pigs (Federation of Animal Science Societies, 2010). Therefore, the increase in summer nursery T_A may have diminished the need for huddling (Hay et al., 2001). Furthermore, this nursery T_A difference may have been responsible for a reduction in eating/drinking and active behavior during the summer in an effort to reduce heat increment from feed consumption during the time of day when behavior was analyzed (Coffey et al., 1982; Nienaber et al., 1999).

As changes in pig behavior can be an indicator of stress and illness, treatment rate with therapeutic antibiotics may be an indicator of illness as well. Therapeutic injectable antibiotics are one of many options currently available to aid in the control of pathogens and disease in addition to good biosecurity practices, vaccinations, and dietary antibiotics (Maes, 2008). In the present study, pigs receiving A had fewer therapeutic antibiotic treatments for enteric challenges compared to GLN pigs during the spring from d 0 to 14 post-weaning, but no differences were detected during the summer. While this may indicate that dietary antibiotic treatments were more effective at reducing pathogen load compared to GLN, the lack of overall dietary treatment differences may suggest that the season of weaning and transport influences the impact of GLN on therapeutic treatments. Regardless, the increase in therapeutic treatments did not appear to coincide with a depression in growth performance and this may be due to differences in the mode of action between A and GLN treatments whereby dietary antibiotics reduce pathogen colonization (Pluske et al., 2002) and GLN fed pigs may improve gut barrier function (Wang et al., 2015). Further work is needed to explore the combined feeding of multiple nutraceuticals that have shown performance benefits independently to determine if the effect of combining them is additive.

In the present study, no dietary treatment carcass trait differences were detected, confirming previous reports that providing dietary antibiotics for a limited period in the nursery phase would have no impact on meat quality (Skinner et al., 2014). While the effects of providing GLN on carcass characteristics in pigs are unknown, previous reports in broilers reported that GLN supplementation during heat stress improves meat yield (Dai et al., 2011). However, because broilers were provided GLN until harvest in the aforementioned study and pigs in the present study were only provided GLN for 14 d, it is likely that the lack of carcass trait differences are related to the timing of dietary inclusion. Nevertheless, a lack of dietary treatment differences confirms that GLN would not have negative effects on carcass traits compared to A and NA diets.

Despite the lack of dietary treatment differences on carcass characteristics, pigs weaned in the spring had greater HCW and loin depth and increased lean percentage and fat-free lean percentage when HCW was used as a covariate compared to summer weaned pigs. While the mechanism(s) for the improvement in carcass characteristics are unknown, we speculate that health status may have impacted the carcass differences observed in the current study due to the differences in therapeutic antibiotic treatment rate between seasons. This response appears to be consistent with previous work by Holck et al., (1998) and Williams et al., (1997) who reported improved carcass characteristics when pigs were reared under higher health status. This suggests that poorer health status may have decreased growth rate and subsequently reduced lean tissue accretion rate. This potential advantage in health status during the spring may have allowed the pigs to grow and deposit lean tissue at a rate closer to their genetic potential because previous studies determined that when pigs were exposed to chronic immune system activation in a health compromised environment, cytokine concentration was elevated (Williams et al., 1997) thereby suppressing lean growth. This is further explained by Zamir et al., (1994) where rats administered with an IL-1 receptor antagonist had reduced skeletal muscle catabolism when IL-1 was administered. Thus, based on these relationships, less environmental pathogens as indicated by reduced therapeutic antibiotic use could have decreased immune system and cytokine activation thus allowing the potential for increased muscle accretion rate due to less skeletal muscle catabolism.

Literature Cited:

Berry R.J., and N.J. Lewis. 2001. The effect of duration and temperature of simulated transport on the performance of early-weaned piglets. Can. J. Anim. Sci. 81:199-204.

Blackshaw, J. K., D. Bodero, and A. W. Blackshaw. 1987. The effect of group composition on behavior and performance of weaned pigs. Appl. Anim. Behav. Sci. 19:73–80.

Bøe, K. 1993. The effect of age at weaning and post-weaning environment on the behaviour of pigs. Acta. Agric. Scand. 43:173-180.

Brooks, P.H., C.A. Moran, J.D. Beal, V. Demeckova, and A. Campbell. 2001. Liquid feeding for the young piglet. In The Weaner Pig: Nutrition and Management, pp. 153–178 [MA Varley and J Wiseman, eds]. Wallingford: CABI Publishing.

Campbell, J. M., J. D. Crenshaw, and J. Polo. 2013. The biological stress of early weaned piglets. J. Anim. Sci. Biotechnol. 4: 19.

Chambers P.G., and T. Grandin. 2001. Guidelines for humane handling, transport and slaughter of livestock. RAP Publication. pp. 33-48.

Chiba, L.I. 2010. Swine production handbook. 13th Revision. Section 6. 62-75.

Coffey, M. T., R. W. Seerley, D. W. Funderburke, and H. C. McCampbell. 1982. Effect of Heat Increment and Level of Dietary Energy and Environmental Temperature on the Performance of Growing-Finishing Swine. J. Anim. Sci. 54:95-105.

Colson, V., E. Martin, P. Orgeur, and A. Prunier. 2012. Influence of housing and social changes on growth, behaviour and cortisol in piglets at weaning. Physiol. Behav. 107:59-64.

Dai, S. F., F. Gaoa, W. H. Zhanga, S. X. Songa, X. L. Xub, and G.H. Zhoub. 2011. Effects of dietary glutamine and gamma-aminobutyric acid on performance, carcass characteristics and serum parameters in broilers under circular heat stress. Anim. Feed Sci. Technol. 168:51-60.

Drake, A., D. Freaser, and D. M. Weary. 2008. Parent–offspring resource allocation in domestic pigs. Behav. Ecol. Sociobiol. 62:309–319.

Dritz, S. S., K. Q. Owen, J. L. Nelssen, R. D. Goodband, and M. D. Tokach. 1996. Influence of Weaning Age and Nursery Diet Complexity on Growth Performance and Carcass Characteristics and Composition of High-Health Status Pigs from Weaning to 109 Kilograms. J. Anim. Sci. 74:2975–2984.

Federation of Animal Science Societies. 2010. Guide for the care and use of agricultural animals in research and teaching. 3rd ed. Fed. Anim. Sci. Soc., Champaign, IL. Chap. 11.

Harris, D.L. 2000. Multi-site pig production. Ames, Iowa. Iowa State University Press.

Hay, M., P. Orgeur, F. Lévy, J. Le Dividich, D. Concordet, R. Nowak, B. Schaal, and P. Morméde. 2001. Neuroendocrine consequences of very early weaning in swine. Physiol. Behav. 72: 263–269.

Hennessy, M. B., T. Deak, and P. A. Schiml-Webb. 2001. Stress-induced sickness behaviors: An alternative hypothesis for responses during maternal separation. Dev. Psychol 39(2):76-83.

Heo, J. M., F. O. Opapeju, J. R. Pluske, J. C. Kim, D. J. Hampson, and C. M. Nyachoti. 2013. Gastrointestinal health and function in weaned pigs: a review of feeding strategies to control post-weaning diarrhoea without using in-feed antimicrobial compounds. 97:207-237.

Hicks, T.A., J.J. McGlone, C.S. Wishant, H.G. Kattesh, and R.L. Norman. 1998. Behavioural, endocrine, immune and performance measures for pigs exposed to acute stress. J. Anim. Sci. 76: 474–483.

Holck, J. T., A. P. Schinckel, J. L. Coleman, V. M. Wilt, M. K. Senn, B. J. Thacker, E. L. Thacker, and A. L. Grant. 1998. The influence of environment on the growth of commercial finisher pigs. Swine Health Prod. 6(4):141-149. Prev. Vet. Med.

Jiang, Z. Y., L. H. Sun, Y. C. Lin, X. Y. Ma, C. T. Zheng, G. L. Zhou, F. Chen, and S. T. Zou. 2009. Effects of dietary glycyl-glutamine on growth performance, small intestinal integrity, and immune responses of weaning piglets challenged with lipopolysaccharide. J. Anim. Sci. 87:4050-4056.

Johnson, J.S., and D.C. Lay Jr. 2017. Evaluating the behavior, growth performance, immune parameters, and intestinal morphology of weaned piglets after simulated transport and heat stress when antibiotics are eliminated from the diet or replaced with L-glutamine. J. Anim. Sci. 95: 91-102.

Lallés, J.P., G. Boudry, C. Favier, N. Le Floc'h, I. Luron, L. Montagne, I.P. Oswald, S. Pié, C. Piel, and B. Séve. 2004. Gut function and dysfunction in young pigs: physiology. Anim. Res. 53: 301–316.

Lambooy, E. 1988. Road transport of pigs over a long distance: some aspects of behavior, temperature and humidity during transport and some effects of the last two factors. Anim. Prod. 46: 257–263.

Lewis, N.J., and Berry R.J., 2006. Effects of season on the behavior of early-weaned piglets during and immediately following transport. Appl. Anim. Behav. Sci. 100: 182-192.

Maenz, D.D., J.F. Patience, and M.S. Wolynetz. 1994. The influence of the mineral level in drinking water and the thermal environment on the performance and intestinal fluid flux of newly weaned pigs. J. Anim. Sci. 72: 300–308.

Maes, D., J. Segales, T. Meyns, M. Sibila, M. Pieters, and F. Haesebrouck. 2008. Control of *Mycoplasma hyopneumoniae* infections in pigs. Vet. Microbiol. 126:297–309.

Meese, G. B. and R. Ewbank. 1973. Establishment and nature of dominance hierarchy in domesticated pigs. Anim. Behav. 21:326–34.

Moeser, A.J., C.V. Klok, K.A. Ryan, J.G. Wooten, D. Little, V.L. Cook, and A.T. Blikslager. 2007. Stress signaling pathways activated by weaning mediate intestinal dysfunction in the pig. Am. J. Physiol. Gastrointest. Liver. Physiol. 292: G173–G181.

National Pork Board. 2015. Transport Quality Assurance Handbook. 5^{th} ed. Natl. Pork Board. Clive, IA.

Nienaber, J. A., G.L. Hahn, and R.A. Eigenberg. 1999. Quantifying livestock responses for heat stress management: a review. Int. J. Biometeorol. 42:183–188.

NRC. 2012. Nutrient requirements of swine. 11^{th} rev. ed. Natl. Acad. Press, Washington, DC.

Peng, X., H. Yan, Z. You, P. Wang, and S. Wang. 2004. Effects of enteral supplementation with glutamine granules on intestinal mucosal barrier function in severe burned patients. Burns 30:135-139.

Pluske, J. R., D. W. Pethick, D. E. Hopwood, and D. J. Hampson. 2002. Nutritional influences on some major enteric bacterial diseases of pigs. Nutr. Res. Rev. 15:333–371.

Schinckel, A. P., J. R. Wagner, J. C. Forrest, and M. E. Einstein. 2010. Evaluation of the prediction of alternative measures of pork carcass composition by three optical probes. J. Anim. Sci. 2010. 88:767–794

Skinner, L. D., C. L. Levesque, D. Wey, M. Rudar, J. Zhu, S. Hooda, and C. F. M. de Lange. 2014. Impact of nursery feeding program on subsequent growth performance, carcass quality, meat quality, and physical and chemical body composition of growing-finishing pigs. J. Anim. Sci. 92:1044-1054.

Smith, M.G., D. Jordan, T.A. Chapman, J.J. Chin, M.D. Barton, T.N. Do, V.A. Fahy, J.M. Fairbrother, and D.J. Trott. 2010. Antimicrobial resistance and virulence gene profiles in multi-drug resistant enterotoxigenic Escherichia coli isolated from pigs with post-weaning diarrhoea. Vet. Microbiol. 145: 299–307.

Van der Meulen, J., S.J. Koopmans, R.A. Dekker, and A. Hoogendoom. 2010. Increasing

weaning age of piglets from 4 to 7 weeks reduces stress, increases post-weaning feed intake but does not improve intestinal functionality. Animal. 4: 1653–1661.
Wamnes, S., N. J. Lewis, and R. J. Berry. 2008. The behaviour of early-weaned piglets following transport: Effect of season and weaning weight. Can. J. Anim. Sci. 88(3): 357-367.
Wang, H., C. Zhang, G. Wu, Y. Sun, B. Wang, B. He. Z. Dai, and Z. Wu. 2015. Glutamine enhances tight junction protein expression and modulates corticotropin-releasing factor signaling in the jejunum of weanling piglets. J. Nutr. 145(1):25-31.
Williams, N. H., T. S. Stahly, and D. R. Zimmerman. 1997. Effect of Level of Chronic Immune System Activation on the Growth and Dietary Lysine Needs of Pigs Fed from 6 to 112 kg. J. Anim. Sci. 75:2481-2496.
Wu, G., S. A. Meier, and D. A. Knabe. 1996. Dietary glutamine supplementation prevents jejunal atrophy in weaned pigs. J. Nutr. 126:2578–2584.
Yi, G. F., J. A. Carroll, G. L. Allee, A. M. Gaines, D. C. Kendall, J. L. Usry, Y. Toride, and S. Izuru. 2005. Effect of glutamine and spray-dried plasma on growth performance, small intestinal morphology, and immune responses of *Escherichia coli* K88$^+$-challenged weaned pigs. J. Anim. Sci. 83:634–643.
Zamir, O., W. O'Brien, R. Thompson, D. C. Bloedow, J. E. Fischer, and P. Hasselgren. 1994. Reduced muscle protein breakdown in septic rats following treatment with interleukin-1 receptor antagonist. Int. J. Biochem. 26:94.

Table 1. Composition of Nursery Diets

	Phase 1[1]			**Phase 2[2]**			**Phase 3[3]**	**Phase 4[4]**
Item	A[5]	GLN[6]	NA[7]	A	GLN	NA		
Ingredient, % as fed								
Corn	30.81	31.18	31.38	37.52	37.89	38.09	51.63	57.38
SBM, 48% CP	13.95	13.95	13.95	18	18	18	25.65	30.7
Dried Distillers Grain with Solubles	---	---	---	---	---	---	---	5
Soybean Oil	5	5	5	5	5	5	3	---
Choice White Grease	---	---	---	---	---	---	---	3
Limestone	0.79	0.79	0.79	0.74	0.74	0.74	0.86	1.33
Monocalcium phosphate	0.4	0.4	0.4	0.49	0.49	0.49	0.49	0.74
Vitamin Premix[8]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Trace Mineral Premix[9]	0.125	0.125	0.125	0.125	0.125	0.125	0.125	0.125
Selenium Premix[10]	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Phytase[11]	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.3	0.35
Plasma Protein	6.5	6.5	6.5	2.5	2.5	2.5	---	---
Spray Dried Blood Meal	1.5	1.5	1.5	1.5	1.5	1.5	---	---
Soy Concentrate	4	4	4	3	3	3	2.5	---
Select Menhaden Fish Meal	5	5	5	4	4	4	4	---
Dried Whey	25	25	25	25	25	25	10	---
Lactose	5	5	5	---	---	---	---	---
Lysine-HCL	0.07	0.07	0.07	0.2	0.2	0.2	0.28	0.4
DL-Methionine	0.22	0.22	0.22	0.23	0.23	0.23	0.18	0.17

L-Threonine	0.04	0.04	0.04	0.09	0.09	0.09	0.12	0.14
L-Tryptophan	---	---	---	0.01	0.01	0.01	0.01	0
Zinc Oxide	0.375	0.375	0.375	0.375	0.375	0.375	0.375	---
Copper Sulphate	---	---	---	---	---	---	---	0.1
Aureomycin 50[12]	0.4	---	---	0.4	---	---	---	---
Denagard 10[13]	0.18	---	---	0.18	---	---	---	---
L-Glutamine[14]	---	0.2	---	---	0.2	---	---	---
Banminth 48[15]	---	---	---	---	---	---	---	0.1
Clarifly, 0.67%[16]	---	---	---	---	---	---	0.08	0.07
Calculated chemical composition								
ME, kcal/kg	3536	3536	3536	3510	3510	3510	3418	3396
Fat, %	7.27	7.27	7.27	7.36	7.36	7.36	5.73	5.86
CP, %	24.62	24.62	24.62	22.87	22.87	22.87	22.29	21.28
SID Lys, %	1.55	1.55	1.55	1.45	1.45	1.45	1.35	1.25
Ca, %	0.9	0.9	0.9	0.85	0.85	0.85	0.8	0.75
Total P, %	0.75	0.75	0.75	0.71	0.71	0.71	0.64	0.57
Avail. P, %	0.6	0.6	0.6	0.55	0.55	0.55	0.45	0.36

[1]Fed d 0 to 7 post-weaning and transport

[2]Fed d 7 to 14 post-weaning and transport

[3]Fed d 14 to 21 post-weaning and transport

[4]Fed d 21 to 34 post-weaning and transport

[5]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)]

[6]Pigs provided 0.20% L-glutamine

[7]Pigs provided no dietary antibiotics

[8]Provided per kilogram of the diet: vitamin A, 6,614 IU; vitamin D3, 661 IU; vitamin E, 44 IU; vitamin K, 2.2 mg; riboflavin, 9 mg; pantothenic acid, 22 mg; niacin, 33 mg

[9]Provided per kilogram of the diet: iron, 289 mg; zinc, 118 mg; manganese, 16 mg; copper, 0.11 mg; iodine, 0.28 mg

[10]Provided 0.3 ppm Se

[11]Provided 600 FTU per kg of the diet

[12]Aureomycin (Zoetis, Parsippany, NJ) provided 441 ppm chlortetracycline in the diet

[13]Denagard (Elanco Animal Health, Greenfield, IN) provided 38.6 ppm tiamulin in the diet

[14]Ajinomoto North America, Inc., Raleigh, NC

[15]Banminth (Phibro Animal Health Corporation, Teaneck, NJ) provided 106 ppm pyrantel tartrate in the diet

[16]Clarifly (Central Life Sciences, Schaumburg, IL) provided 5.4 ppm (Phase 3) and 4.7 ppm (Phase 4) diflubenzuron in the diet

Table 2. Ethogram used for behavioral observations.

Category	Behavior	Definition
Sickness Behavior	Huddling	When 3 or more pigs are touching while lying down and 50% of a pig's body is touching another pig.
Other	Active	Piglets are walking about or interacting in a non-aggressive manner with each other or their environment.
	Resting	Piglets are lying, either ventral or lateral, either alone or loosely in groups, with gaps of spaces between them.
	Aggressive	Piglets are engaged in agonistic interactions.
	Eating/Drinking	The piglet has its nose in the feeder or its mouth on the waterer.
	Non-visible	When piglet moves out of view and cannot be observed.

Table 3. Composition of grow-finish diets.

Item	Phase 1[1]	Phase 2[2]	Phase 3[3]	Phase 4[4]	Phase 5[5]	Phase 6[6]
Ingredient, % as fed						
Corn	61.47	64.65	66.40	71.10	82.38	68.67
SBM, 48% CP	23.20	16.15	9.75	5.25	4.25	15.10
Dried Distillers Grain with Solubles	10.00	15.00	20.00	20.00	10.00	10.00
Choice White Grease	2.00	1.00	1.00	1.00	1.00	3.00
Limestone	1.37	1.35	1.39	1.32	1.16	1.26
Monocalcium phosphate	0.47	0.32	0.05	0.00	0.10	0.27
Vitamin Premix	0.150[7]	0.150[7]	0.125[8]	0.120[9]	0.100[10]	0.150[7]
Trace Mineral Premix	0.10[11]	0.09[12]	0.08[13]	0.07[14]	0.05[15]	0.10[11]
Selenium Premix	0.050[16]	0.050[16]	0.050[16]	0.050[16]	0.025[17]	0.050[16]
Phytase[18]	0.10	0.10	0.10	0.10	0.10	0.10
Salt	0.35	0.35	0.30	0.30	0.25	0.30
Lysine-HCL	0.42	0.46	0.48	0.46	0.37	0.42
DL-Methionine	0.11	0.08	0.05	0.01	0.00	0.10
L-Threonine	0.130	0.130	0.120	0.105	0.095	0.160
L-Tryptophan	0.010	0.030	0.035	0.040	0.030	0.030
Paylean 2.25[19]	---	---	---	---	---	0.15
Availa Zn 120[20]	---	---	---	---	---	0.042
Clarifly, 0.67%	0.07[21]	0.09[22]	0.07[21]	0.08[23]	0.09[22]	0.10[24]
Calculated chemical composition						
ME, kcal/kg	3373	3337	3351	3359	3371	3438
Fat, %	5.29	4.69	5.06	5.15	4.73	6.40
CP, %	19.34	17.59	15.99	14.18	11.90	16.01
SID Lys, %	1.10	0.98	0.85	0.73	0.60	0.90
Ca, %	0.70	0.65	0.60	0.55	0.50	0.60
Total P, %	0.50	0.47	0.41	0.38	0.35	0.42
Avail. P, %	0.32	0.30	0.26	0.24	0.20	0.26

[1]Fed d 0 to 21 of the grow-finish phase

[2]Fed d 21 to 42 of the grow-finish phase

[3]Fed d 42 to 62 of the grow-finish phase

[4]Fed d 62 to 83 of the grow-finish phase

[5]Fed d 83 to 104 of the grow-finish phase

[6]Fed d 104 to 125 of the grow-finish phase

[7]Provided per kilogram of the diet: vitamin A, 3,968 IU; vitamin D3, 397 IU; vitamin E, 26 IU; vitamin K, 1.3 mg; riboflavin, 5 mg; pantothenic acid, 13 mg; niacin, 20 mg

[8]Provided per kilogram of the diet: vitamin A, 3,307 IU; vitamin D3, 331 IU; vitamin E, 22 IU; vitamin K, 1.1 mg; riboflavin, 4 mg; pantothenic acid, 11 mg; niacin, 17 mg

[9]Provided per kilogram of the diet: vitamin A, 3,175 IU; vitamin D3, 317 IU; vitamin E, 21 IU; vitamin K, 1.1 mg; riboflavin, 4 mg; pantothenic acid, 11 mg; niacin, 16 mg
[10]Provided per kilogram of the diet: vitamin A, 2,646 IU; vitamin D3, 265 IU; vitamin E, 18 IU; vitamin K, 0.9 mg; riboflavin, 4 mg; pantothenic acid, 9 mg; niacin, 13 mg
[11]Provided per kilogram of the diet: iron, 232 mg; zinc, 94 mg; manganese, 13 mg; copper, 0.09 mg; iodine, 0.23 mg
[12]Provided per kilogram of the diet: iron, 208 mg; zinc, 85 mg; manganese, 12 mg; copper, 0.08 mg; iodine, 0.20 mg
[13]Provided per kilogram of the diet: iron, 185 mg; zinc, 75 mg; manganese, 10 mg; copper, 0.07 mg; iodine, 0.18 mg
[14]Provided per kilogram of the diet: iron, 162 mg; zinc, 66 mg; manganese, 9 mg; copper, 0.06 mg; iodine, 0.16 mg
[15]Provided per kilogram of the diet: iron, 116 mg; zinc, 47 mg; manganese, 6 mg; copper, 0.05 mg; iodine, 0.11 mg
[16]Provided 0.3 ppm Se
[17]Provided 0.15 ppm Se
[18]Provided 600 FTU per kg of the diet
[19]Paylean (Elanco Animal Health, Greenfield, IN) provided 7.5 ppm ractopamine HCl in the diet
[20]Zinpro Corporation, Eden Prairie, MN
[21]Clarifly (Central Life Sciences, Schaumburg, IL) provided 4.7 ppm diflubenzuron in the diet
[22]Clarifly (Central Life Sciences, Schaumburg, IL) provided 6.0 ppm diflubenzuron in the diet
[23]Clarifly (Central Life Sciences, Schaumburg, IL) provided 5.4 ppm diflubenzuron in the diet
[24]Clarifly (Central Life Sciences, Schaumburg, IL) provided 6.7 ppm diflubenzuron in the diet

Table 4. Effect of dietary treatment and season on nursery pig growth performance.[1]

	Main Effects								
	Season		Diet				*P*		
Parameter	Summer[2]	Spring[3]	A[4]	GLN[5]	NA[6]	SE	D[7]	S[8]	D x S
Day 0 to 14									
Initial BW, kg	5.64	5.51	5.58	5.59	5.57	0.29	0.99	0.70	0.99
ADG, g	210.09	206.08	224.23[a]	210.78[a]	189.24[b]	10.19	0.01	0.56	0.82
ADFI, g	274.57	260.62	277.10[a]	272.06[ab]	253.63[b]	13.21	0.04	0.08	0.92
G:F	0.80	0.80	0.84[a]	0.79[b]	0.77[b]	0.01	0.01	0.91	0.17
Day 14 BW, kg	8.44	8.46	8.65[a]	8.50[a]	8.19[b]	0.52	0.01	0.83	0.97
Day 14 to 34									
ADG, g	439.00	455.71	458.01	447.36	436.69	12.05	0.21	0.09	0.43
ADFI, g	693.52	674.65	702.26	680.43	669.56	22.81	0.16	0.19	0.63
G:F	0.63	0.68	0.65	0.66	0.65	0.01	0.78	0.01	0.04
Day 0 to 34									
ADG, g	347.43	355.85	364.50[a]	352.73[ab]	337.71[b]	10.18	0.01	0.23	0.58
ADFI, g	525.94	509.04	532.20[x]	517.08[xy]	503.19[y]	17.43	0.09	0.12	0.77
G:F	0.70	0.73	0.73[a]	0.71[ab]	0.70[b]	0.01	0.03	0.01	0.07
Final BW, kg	17.20	17.62	17.78[a]	17.49[ab]	16.96[b]	0.74	0.04	0.11	0.69

[1]A total of 10 pens were used per dietary treatment per season

[2]Pigs weaned and transported for 12 h during July 2016

[3]Pigs weaned and transported for 12 h during April 2017

[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets

[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets

[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets

[7]Dietary treatment

[8]Season

[a,b]Letters indicate significant differences ($P \leq 0.05$) within a row and main effect

[x,y]Letters indicate tendencies ($0.05 < P \leq 0.10$) within a row and main effect

Table 5. Effect of dietary treatment and season on grow-finish pig growth performance.[1]

	Main Effects								
	Season		Diet				*P*		
Parameter	Summer[2]	Spring[3]	A[4]	GLN[5]	NA[6]	SE	D[7]	S[8]	D x S
Day 0 to 62									
ADG, kg	0.76	0.77	0.78	0.76	0.76	0.01	0.32	0.37	0.62
ADFI, kg	1.79	1.75	1.80	1.76	1.75	0.03	0.40	0.14	0.88
G:F	0.44	0.46	0.45	0.46	0.45	0.01	0.80	0.01	0.36
Day 62 BW, kg	64.72	65.50	65.99	65.02	64.31	0.96	0.22	0.32	0.76
Day 62 to 125									
ADG, kg	0.82	0.96	0.88	0.89	0.90	0.02	0.41	0.01	0.36
ADFI, kg	2.83	2.96	2.87	2.91	2.90	0.05	0.72	0.01	0.42
G:F	0.29	0.33	0.30	0.31	0.31	0.01	0.17	0.01	0.62
Day 0 to 125									
ADG, kg	0.79	0.87	0.83	0.83	0.83	0.01	0.95	0.01	0.58
ADFI, kg	2.31	2.35	2.33	2.33	2.32	0.03	0.97	0.21	0.60
G:F	0.37	0.39	0.38	0.38	0.38	0.01	0.54	0.01	0.56
Final BW, kg	117.37	127.19	122.77	121.73	122.34	1.23	0.83	0.01	0.64

[1]A total of 10 pens were used per dietary treatment per season

[2]Pigs weaned and transported for 12 h during July 2016

[3]Pigs weaned and transported for 12 h during April 2017

[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets

[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets

[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets

[7]Dietary treatment

[8]Season

Table 6. Effect of season and dietary treatment on behavior (% of time) during the nursery period.[1]

	Main Effects								
	Season		Diet				*P*		
Behavior	Summer[2]	Spring[3]	A[4]	GLN[5]	NA[6]	SE	D[7]	S[8]	D x S
Day 2 to 12 post-weaning									
Huddling[9], %	5.52	15.38	10.30	8.58	11.20	1.46	0.92	<0.01	0.84
Active[10], %	9.14	12.49	10.90	10.64	10.71	0.55	0.78	<0.01	0.14
Resting[11], %	77.55	73.07	73.6	77.13	74.94	1.33	0.12	0.34	0.33
Aggressive[12], %	1.39	1.57	1.74[x]	1.28[y]	1.41[xy]	0.19	0.09	0.14	0.70
Eat/Drink[13], %	8.70	11.26	10.70	9.96	9.14	0.51	0.17	<0.01	0.18
Non-visible[14], %	0.75	0.34	0.83	0.41	0.36	0.37	0.26	0.04	0.67

[1]A total of 10 pens were used per dietary treatment per season

[2]Pigs weaned and transported for 12 h during July 2016

[3]Pigs weaned and transported for 12 h during April 2017

[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets

[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets

[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets

[7]Dietary treatment

[8]Season

[9]When 3 or more pigs are touching while lying down and 50% of a pig's body is touching another pig; collected independent of other behaviors

[10]Piglets are walking about or interacting in a non-aggressive manner with each other or their environment

[11]Piglets are lying, either ventral or sternal, either alone or loosely in groups, with gaps of spaces between them

[12]Piglets are engaged in agonistic interactions

[13]The piglet has its nose in the feeder or its mouth on the waterer

[14]When piglet moves out of view and cannot be observed

[x,y]Letters indicate tendencies ($0.05 < P \leq 0.10$) within a row

Table 7. Effect of dietary treatment and season on therapeutic antibiotic treatment rate during the nursery period.[1]

	Main Effects									
	Summer[2]			Spring[3]				*P*		
Parameter	A[4]	GLN[5]	NA[6]	A	GLN	NA	SE	D[7]	S[8]	D x S
Day 0 to 14										
Enteric[9]	3.13[y]	3.75[y]	6.88[xy]	3.13[y]	8.19[x]	4.55[xy]	2.31	0.31	0.38	0.07
Lame[10]	1.88	1.88	1.25	0.63	1.39	0.63	1.02	0.73	0.27	0.89
Unthrifty[11]	1.88	1.25	1.25	0.00	0.69	1.25	1.02	0.92	0.22	0.48
Respiratory[12]	---	---	---	---	---	---	---	---	---	---
Other[13]	0.00	0.00	0.00	1.25	0.69	1.25	0.86	0.86	0.02	0.86
Day 14 to 34										
Enteric	0.00	0.95	0.48	0.00	0.00	0.56	0.66	0.36	0.33	0.37
Lame	0.48[b]	0.00[b]	0.00[b]	0.56[b]	2.12[a]	0.00[b]	1.00	0.08	0.06	0.04
Unthrifty	0.00	0.00	1.03	1.03	1.15	1.03	0.80	0.64	0.14	0.57
Respiratory	0.00	0.00	0.48	0.00	0.53	0.00	0.53	0.58	0.94	0.20
Other	0.00	0.00	0.00	0.00	0.53	0.00	0.53	0.31	0.27	0.31
Day 0 to 34										
Enteric	1.25	2.07	3.04	1.25	3.17	2.15	1.05	0.15	0.64	0.24
Lame	1.04	0.75	0.50	0.58	1.83	0.25	0.70	0.14	0.83	0.20
Unthrifty	0.75	0.50	1.12	0.62	0.97	1.12	0.55	0.63	0.88	0.79
Respiratory	0.00	0.00	0.29	0.00	0.32	0.00	0.32	0.58	0.94	0.20
Other	0.00	0.00	0.00	0.50	0.60	0.50	0.42	0.99	0.01	0.99

[1]A total of 10 pens were used per dietary treatment per season

[2]Pigs weaned and transported for 12 h during July 2016

[3]Pigs weaned and transported for 12 h during April 2017

[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets

[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets

[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets

[7]Dietary treatment

[8]Season

[9]Percent of pigs within pen treated with therapeutic antibiotics for enteric challenge

[10]Percent of pigs within pen treated with therapeutic antibiotics for lameness

[11]Percent of pigs within pen treated with therapeutic antibiotics for un-thriftiness

[12]Percent of pigs within pen treated with therapeutic antibiotics for respiratory challenge

[13]Percent of pigs within pen treated with therapeutic antibiotics for all other conditions

[a,b,c]Letters indicate significant differences ($P \leq 0.05$) within a row

[x,y]Letters indicate tendencies ($0.05 < P \leq 0.10$) within a row

Table 8. Effect of season and dietary treatment on therapeutic antibiotic treatment rate for enteric challenges during the grow-finish period.[1]

Parameter	Summer[2] A[4]	Summer[2] GLN[5]	Summer[2] NA[6]	Spring[3] A	Spring[3] GLN	Spring[3] NA	SE	***P*** D[7]	***P*** S[8]	***P*** D x S
D 0 to 62										
Enteric[9]	0.56	0.00	0.00	0.56	0.67	0.67	0.67	0.81	0.31	0.77
Lame[10]	0.56	0.56	1.89	0.00	0.00	0.00	1.06	0.41	0.02	0.41
Unthrifty[11]	1.78	0.00	0.67	0.00	0.00	0.56	0.99	0.24	0.17	0.16
Respiratory[12]	11.11	10.78	8.00	0.56	1.11	2.22	2.92	0.60	<0.01	0.77
Other[13]	1.11	2.33	1.33	0.00	0.00	0.56	1.11	0.69	0.02	0.45
D 62 to 125										
Enteric	0.00	0.67	0.00	0.00	1.67	0.67	0.93	0.08	0.19	0.58
Lame	2.22	0.67	0.67	0.00	0.00	0.00	1.05	0.21	0.01	0.21
Unthrifty	0.56	0.00	0.00	1.67	0.00	0.00	0.93	0.01	0.28	0.30
Respiratory	10.33	8.22	12.17	7.33	7.39	6.78	4.20	0.81	0.49	0.86
Other	0.56	0.00	0.00	0.00	0.00	0.00	0.56	0.37	0.32	0.37
D 0 to 125										
Enteric	0.28	0.33	0.00	0.28	1.17	0.67	0.57	0.37	0.10	0.47
Lame	1.39	0.61	1.28	0.00	0.00	0.00	0.62	0.52	<0.01	0.52
Unthrifty	1.17	0.00	0.33	0.83	0.00	0.28	0.57	0.01	0.70	0.88
Respiratory	10.72	9.50	10.08	3.94	4.25	4.50	2.35	0.90	<0.01	0.95
Other	0.83	1.17	0.67	0.00	0.00	0.28	0.57	0.90	0.01	0.50

[1]A total of 10 pens were used per dietary treatment per season
[2]Pigs weaned and transported for 12 h during July 2016
[3]Pigs weaned and transported for 12 h during April 2017
[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets
[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets
[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets
[7]Dietary treatment
[8]Season
[9]Percent of pigs within pen treated with therapeutic antibiotics for enteric challenge
[10]Percent of pigs within pen treated with therapeutic antibiotics for lameness
[11]Percent of pigs within pen treated with therapeutic antibiotics for un-thriftiness
[12]Percent of pigs within pen treated with therapeutic antibiotics for respiratory challenge
[13]Percent of pigs within pen treated with therapeutic antibiotics for all other conditions

Table 9. Effect of season and dietary treatment on carcass characteristics.[1]

	Main Effects								
	Season		**Diet**				***P***		
Parameter	Summer[2]	Spring[3]	A[4]	GLN[5]	NA[6]	SE	D[7]	S[8]	D x S

No HCW[9] covariate									
HCW, kg	92.42	97.44	95.32	95.54	93.93	1.32	0.60	<0.01	0.70
Loin Depth, mm	63.95	67.46	65.79	65.85	65.48	0.72	0.93	<0.01	0.60
Backfat, mm	21.35	22.05	21.73	21.64	21.73	0.59	0.99	0.31	0.40
Yield, %	77.18	75.67	76.55	76.36	76.36	0.19	0.68	<0.01	0.46
Lean, %[10]	54.42	54.61	54.51	54.55	54.47	0.25	0.97	0.53	0.54
Fat-free lean, %[11]	48.69	48.79	48.74	48.79	48.69	0.30	0.97	0.76	0.50
HCW covariate									
Loin Depth, mm	64.43	66.99	65.72	65.74	65.68	0.69	0.99	0.01	0.66
Backfat, mm	22.02	21.41	21.64	21.49	22.00	0.49	0.75	0.33	0.57
Yield, %	77.33	75.52	76.52	76.33	76.42	0.17	0.69	0.01	0.63
Lean, %	54.20	54.82	54.54	54.60	54.38	0.23	0.78	0.04	0.71
Fat-free lean, %	48.41	49.06	48.77	48.85	48.58	0.27	0.77	0.07	0.68

[1]A total of 10 pens were used per dietary treatment per season

[2]Pigs weaned and transported for 12 h during July 2016

[3]Pigs weaned and transported for 12 h during April 2017

[4]Pigs provided dietary antibiotics [chlortetracycline (441 ppm) + tiamulin (38.6 ppm)] for 14 d post-weaning and transport and then fed common antibiotic free diets

[5]Pigs provided 0.20% L-glutamine for 14 d post-weaning and transport and then fed common antibiotic free diets

[6]Pigs provided no dietary antibiotics for 14 d post-weaning and transport and then fed common antibiotic free diets

[7]Dietary treatment

[8]Season

[9]Hot carcass weight

[10]Equation used: 54.672154 - (0.412525 × backfat, mm) - (0.002982 × hot carcass weight, kg × 2.20462) + (0.1433242 × loin depth, mm) (Indiana Packers Corporation, 2015)

[11]Equation used: 51.2 - (0.510 × backfat, mm) + (0.131 × loin depth, mm) (Schinckel et al., 2010)

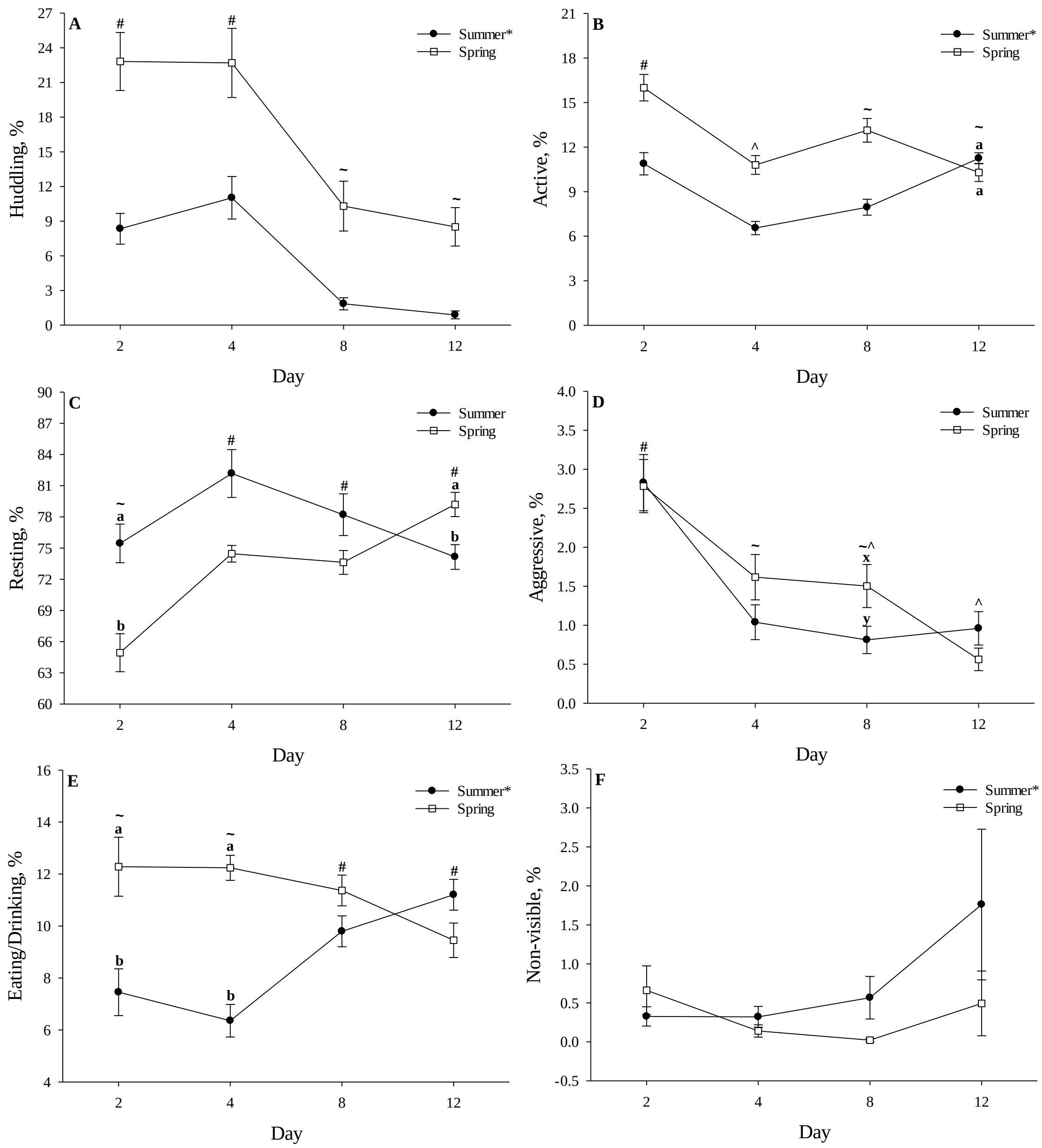

Figure 1: The effects of season (summer or spring) on (A) huddling, (B) active, (C) resting, (D) aggressive, (E) eating/drinking, and (F) non-visible behavior on d 2, 4, 8, and 12 post-weaning and transport. An asterisk (*) on the legend indicates overall season differences ($P < 0.05$), #,~,^ symbols indicate overall day differences ($P < 0.05$), ^{a,b}letters indicate season by day differences ($P \leq 0.05$), and ^{x,y}letters indicate season by day tendencies ($0.05 < P \leq 0.10$).