RESEARCH REPORT
ANIMAL WELFARE

Title: Long distance transport of breeding stock- **NPB #06-181**

Investigator: John J. McGlone

Institution: Texas Tech University / Pork Industry Institute

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Industry Summary:
Breeding stock are frequently shipped considerable distance from isolated high-level biosecurity facilities to farms across North America. Little scientific data exists on the reproductive or health effects of long-distance transportation on breeding stock. To address this lack of knowledge we shipped breeding-aged gilts at various space allowances for different lengths of time up to 30 hours.

During the first experiment we shipped pigs for 30 hours at TQA recommended space allowances plus an extra 20%, which is common practice when shipping breeding gilts. Every 6 hours we assessed health and well-being of the gilts by measuring weight loss, dehydration, stress levels, and blood chemistry parameters. During the second experiment we compared 2 space allowances, TQA recommended space allowance and TQA +20% recommended space allowance. In each study a group of gilts remained in their home pen to serve as controls. At the end of each study gilts were returned to the TTU herd as replacements and their reproductive performance was assessed.

In both studies a similar pattern in the physiological response to transport was observed in the gilts. An initial negative response to transport was followed by an adaption period, and then as the transport period increased a gradual increase in the number of physiological changes that differed from control gilt was observed. Space allowance (TQA and TQA +20%) did not affect any of the measured variables. After a 6 and 12 hour transport period there were significant changes in cortisol concentrations, neutrophil to lymphocyte ratio, and glucose concentrations. These changes suggest an acute stress response to transportation at 6 and 12 hours. After a 6 hour transport period total protein and weight loss were significantly different compared with control gilts, suggesting that transported pigs were experiencing dehydration. Changes in aspartate aminotransferase, creatine kinase, and blood urea nitrogen concentrations at transport durations longer than 6 hours indicate increasing fatigue as a response to physical activity and possibly tissue damage. Reproductive performance (measured as farrowing rate, total born, born alive, stillborn, mummies, weight at processing, number weaned, and weight at weaning) was not affected by transport or duration of transportation in either of the studies.

These data support the growing body of evidence showing that short duration transportation can be stressful to pigs but some degree of acclimation occurs as transport continues. However, after a 30 hour transport period some variables indicative of animal well-being appeared to decrease possibly due to dehydration and hunger. Even though physiological changes were observed in gilts in response to transport, these physiological effects appeared to be only transitory as there was no effect of transport or transport duration on the reproductive performance of these breeding gilts. When carefully managed, healthy gilts can travel long distances with little risk of long term effects on health, well-being, and reproductive performance.

Scientific Abstract:
Long duration transport is an important animal welfare issue. Little experimental work exists on the effects of long duration transportation on breeding gilts. The goal of this project was to evaluate the effects of a 30 hour transport on acute physiological measures and reproductive parameters in breeding aged gilts.

In experiment 1, gilts were transported for up to 30 hour in a trailer allowing a space allowance of 0.504 m^2/head which is common practice when shipping breeding gilts. In addition to 10 gilts per experimental treatment on the trailer, 3 control gilts remained in the home pen. Every 6 hours, pigs from one compartment were removed from the trailer (6, 12, 18, 24, and 30 hours duration of transport). Blood samples and body weights were collected from groups of gilts and their respective controls before and after transport (at 0, 6, 12, 18, 24, and 30 hours). The experiment was repeated twice in July 2007 in Lubbock, TX, USA and included 120 pigs. Leukocyte numbers and percentages, platelets, cortisol, albumin, aspartate aminotransferase, bilirubin, blood urea nitrogen, creatine kinase, glucose, total protein, and interleukin-8 concentrations differed in transported compared to control gilts. Cortisol and glucose concentrations, and the N:L, which are indicative of acute stress response to transportation, decreased as transportation duration increased. Total protein concentrations, indicative of mild dehydration, decreased after a 6 hour transport period. Reproductive performance (measured as farrowing rate, total born, born alive, stillborn, mummies, weight at processing, number weaned, and weight at weaning) was not affected by transport or duration of transportation. By following TQA guidelines, it is possible to transport breeding gilts in hot weather with limited short-term physiological effects and no long-term effects on health or reproductive performance.

In experiment 2, 2 space allowances were replicated 5 times in a commercial semi-trailer that allowed 0.334 and 0.409 m^2/pig. In addition to 4 gilts per experimental treatment on the trailer, 4 control gilts remained in the home pen. Every 6 hours, pigs in one pair of compartments were removed from the trailer (6, 12, 18, 24, and 30 hours duration of transport). Blood samples and body weights were collected from groups of gilts and their respective controls before and after transport (at -24, 6, 12, 18, 24, and 30 hours). The experiment was repeated twice in October 2007 in Lubbock, TX, USA and included 120 pigs. Weight loss was greater ($P< 0.05$) among transported pigs relative to controls after 6 hours of transport, however, after this initial weight loss body weights did not differ ($P> 0.05$) between transported and control gilts from 12 through to 30 hours of transport. Additionally, space allowance did not affect ($P> 0.05$) weight loss in gilts over time. Transient changes were seen in glucose, total protein, albumin, lymphocyte numbers, granulocytes numbers, neutrophil:lymphocyte ratio, hematocrit, and platelet numbers. Cortisol, creatine kinase, and aspartate aminotransferase differed from control gilts but not between space allowances. Reproductive performance (measured as farrowing rate, total born, born alive, stillborn, mummies, weight at processing, number weaned, and weight at weaning) was not affected by transport or duration of transportation.

The results from experiment 1 and 2 suggest that gilts transported for up to 30 hours only experience transient changes in physiological measures of stress as assessed by the parameters evaluated in these studies. Overall, these data indicate that the 28-hour law may be too conservative as we found no overwhelming negative health or well-being effects on breeding gilts after 30 hours of transport as compared to non-transported control gilts.

Introduction:

Breeding stock in the USA and Canada are transported for many hours and sometimes days. While transport losses are not common, at times significant transport losses have been observed on long hauls. Although the percentage of pig losses during transport is low, it is of economic concern for producers and also a significant welfare concern. The majority of research on pig transport has focused on the welfare of slaughter weight pigs during transport to the processing plant. However, transport of breeding stock is an important welfare issue as these animals are often transported for over 12 hours in the US.

The majority of pigs being transported occur when they reach market weight and are transported by truck from the farm to the processing plant. Most pigs arrive at the processing plant and are processed without incident. However, a small percentage of pigs are dead or injured at or prior to arrival at the processing plant. The cause of dead and injured pigs during transport is not well understood (Ellis et al., 2003). Speculation into the factors that affect the percentage of dead and injured pigs during transport include genetics (Ellis et al., 2003), handling of pigs prior to and after transport (Peeters et al., 2004), and perhaps the overall stress caused by mixing pigs with conspecifics and exposure to a novel environment. Other factors that may be involved in the percentage of dead and injured pigs during transport include factors directly associated with transport including air temperature, relative humidity, travel time, and availability of water and food.

Few studies have reported the welfare of pigs under commercial conditions directly related to factors involved in long distance transport of breeding stock. Transport stress has been shown to influence several physiological and behavioral measures in pigs. Furthermore, sub-optimal transport conditions can reduce the welfare and health of pigs. Therefore, transporting pigs' long distances is not only an animal health issue but a welfare concern for the swine industry. To date there is very little information available on the effects of long periods of transport on the welfare of breeding stock. To improve our knowledge and understanding of this subject and potentially the welfare of the pigs more information is needed on the effects of transport on breeding gilts throughout a range of seasons by using a multi-disciplinary approach including animal performance, physiology, and behavior.

Objectives:

The objective of this research project is to determine the effects of transport and season on pig welfare (performance, physiology, behavior, and injury) during transport of breeding stock being transported for 30 hours.

Materials and Methods:

EXPERIMENT 1 (Summer)

Animals

Breeding gilts (Camborough-22) were obtained from a PIC multiplier site and transported to the Texas Tech University swine farm in New Deal, TX. Upon arrival the gilts were approximately 9-10 weeks old and 25 kg. Gilts were raised to a weight of 123 kg prior to the commencement of experiment 1. All animals were fed a diet to meet or exceed NRC nutrient requirements. Water was provided ad libitum. All animal procedures were approved by the Texas Tech University Animal Care and Use Committee.

This experiment was designed to evaluate two aspects of long distance transport: 1) the immediate physiological response of breeding gilts to long distance transport, and 2) the reproductive performance of breeding age gilts exposed to a long distance transport.

Trailer design

An 11 m gooseneck trailer was modified to create three compartments (2.1 × 2.4 m). Each compartment held 10 gilts creating a space allowance of 0.504 m^2/head. This space allowance was the trucking quality assurance (TQA) recommended space allowance for pigs this size plus 20%. A space allowance of TQA plus 20 % was used as this is the space allowance at which breeding gilts are commonly transport at. The trailer floor consisted of slatted wood. Straw was used in the trailer during transport. The trailer was pulled by a series of drivers, each driving one six-hour shift. The same drivers were used for each of the two replicates.

Experimental design

The study was conducted over July, 2007. The maximum-minimum temperature range during the first replicate was 18.3-38.9°C (65-102°F) and 18.8-37.2°C (66-99°F) for the second replicate. The maximum-minimum humidity range during the first replicate was 24-95% and 25-85% for the second replicate. Ten weight matched gilts were allocated to one of 6 transport treatment groups; 6, 12, 18, 24, and 30 hours. In addition to the 10 gilts per experimental treatment on the trailer, 3 control gilts remained in their home pen. Every 6 h, pigs in one compartments were removed from the trailer (6, 12, 18, 24, and 30 h duration of transport). Blood samples were collected from transported gilts and their respective controls before and after transport (at -24, 6, 12, 18, 24, and 30h). The experiment was repeated twice.

Blood analysis

Ten mL of whole blood was collected via jugular veinapuncture into heparinized tubes before and after transport. Whole blood was analyzed to determine white and red blood cell counts, differential leukocyte counts, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentrations, red blood cell distribution width, and platelet counts using a cell counter (Cell-Dyn® 1800, Abbott laboratories, Abbott Park, IL) and the granulocyte to lymphocyte (N:L) ratio was calculated by dividing the percent of granulocytes by the percent of lymphocytes. Plasma was collected, frozen, and stored at -20 °C. Plasma was sent to the Iowa State University Veterinary Pathology Laboratory (Ames, IA) to determine metabolism measures including blood urea nitrogen (BUN), creatinine, glucose, total protein, albumin, aspartate aminotransferase (AST), creatine kinase (CK), alkaline phosphatase (Alk Phos), gamma glutamyltransferase (GGT), total bilirubin, hemolytic index, and lipemic index. Plasma was analyzed for cortisol concentration using a commercially available enzyme Immunoassay kit (Assay Designs, Ann Harbor, MI). A sub-set of plasma samples were analyzed for the cytokines IL-1β, IL-2, IL-4, IL-6, IL-8, IL-10, IL-12p70, and IFN-γ using a commercially available kit (Searchlight porcine cytokine kit (Pierce, Rockford, IL).

Reproductive performance

Staff at the university farm began including the replacement gilts into their breeding program as soon as the experiments were over. The conditions at the university farm differ from those found at commercial facilities, and as such these data need to be interpreted with reference to the control treatment and not industry averages. Farrowing rate, total born, born alive, stillborn, processing weight, number weaned, and weaning weight were all calculated for first parities.

EXPERIMENT 2 (Fall)

Animals

Breeding gilts (Camborough-22) were obtained from a PIC multiplier site and transported to the Texas Tech University swine farm in New Deal, TX. Upon arrival the gilts were approximately 9-10 weeks old and 25 kg. Gilts were raised to a weight of 80 kg prior to the commencement of experiment 2. All animals were fed a diet to meet or exceed NRC nutrient requirements. Water was

provided ad libitum. All animal procedures were approved by the Texas Tech University Animal Care and Use Committee.

This experiment was designed to evaluate two aspects of long distance transport: 1) the immediate physiological response of breeding gilts to long distance transport, and 2) the reproductive performance of breeding age gilts exposed to a long distance transport.

Trailer design

A Wilson straight-deck livestock trailer (16.2 m) was modified to create ten compartments. Only the bottom deck of the trailer was utilized for this experiment. Two space allowances were compared in this study, TQA and TQA plus 20% (1.09 x 1.22 m and 1.34 x 1.22m, respectively). The two space allowances were replicated 5 times. Each compartment held 4 gilts, creating a space allowance of 0.334 m2/head and 0.409 m2/head, representing TQA and TQA plus 20% respectively. The trailer floor was solid aluminum with embossed tread and covered with wood shavings during transport.

Experimental design

The study was conducted over October, 2007. Four weight matched (Control 89.5 ± 1.4 kg, TQA 91.9 ±1.4 kg, TQA +20% 92.7 ±1.5 kg) gilts were allocated to one of 12 transport times and TQA or TQA plus space allowance; 6, 12, 18, 24, and 30 hours. In addition to the four gilts per experimental treatment on the trailer, four control gilts remained in their home pen. Every 6 h, pigs in one pair of compartments (TQA and TQA plus) were removed from the trailer (6, 12, 18, 24, and 30 h duration of transport). Blood samples were collected from transported gilts and their respective controls before and after transport (at -24, 6, 12, 18, 24, and 30h). Control gilts were bled at the same times as their respective experimental group. The experiment was repeated twice. Gilt body weights were recorded prior to the start of the trial and as soon as the gilts came off of the trailer. Control gilts were weighed at the same times.

Blood analysis

Ten mL of whole blood was collected via jugular veinapuncture into heparinized tubes before and after transport. Whole blood was analyzed to determine white blood cell counts, differential leukocyte counts, hemoglobin, hematocrit, and platelet counts using a cell counter (Cell-Dyn® 1800, Abbott laboratories, Abbott Park, IL) and the granulocyte to lymphocyte (N:L) ratio was calculated by dividing the percent of granulocytes by the percent of lymphocytes. Plasma was collected, frozen, and stored at -20 °C. Plasma was sent to the Iowa State University Veterinary Pathology Laboratory (Ames, IA) to determine metabolism measures including blood urea nitrogen (BUN), creatinine, glucose, total protein, albumin, aspartate aminotransferase (AST), creatine kinase (CK), alkaline phosphatase (Alk Phos), gamma glutamyltransferase (GGT), total bilirubin, hemolytic index, and lipemic index. Plasma was analyzed for cortisol concentration using a commercially available enzyme Immunoassay kit (Assay Designs, Ann Harbor, MI).

Reproductive performance

Staff at the university farm began including the replacement gilts into their breeding program as soon as the experiments were over. The conditions at the university farm differ from those found at commercial facilities, and as such these data need to be interpreted with reference to the control treatment and not industry averages. Farrowing rate, total born, born alive, stillborn, processing weight, number weaned, and weaning weight were all calculated for first parities.

Statistical analysis

Data that did not meet the assumptions of normality and homoscedasticity were log transformed prior to statistical analysis. Data that still did not meet those assumptions were analyzed with non-parametric tests. For the hematological and blood chemistry variables ANCOVAs were run

with PROC GLM in SAS (SAS Inst. Inc., Cary, NC). The baseline blood values were used as covariates for each individual. The model included the effects of time, treatment (control or transported in summer; control, TQA, or TQA+20% in fall), replicate, and a two-way interaction (time*treatment). Most reproductive variables and cytokines were analyzed with ANOVA using PROC GLM in SAS. A 2 x 5 contingency table (Chi-Square distribution) was used for farrowing rate using PROC FREQ in SAS. Stillborn and born alive were analyzed by Kruskall-Wallis in SAS as PROC NPAR1WAY. Least square means were evaluated using the PDIFF option of SAS. Graphs and tables display real data (not transformed) summarized by means ±SE. Statistical significance was set at $p< 0.05$.

Results:

EXPERIMENT 1 (Summer)

Whole blood hematology

The total white blood cell count, lymphocytes counts and percent lymphocytes, N:L ratio, and platelet count differed ($p<0.05$) between treatments at different transport times (Table 4). (Lymphocytes counts: $F= 7.21$; d.f.= 5, 195; p-value< 0.0001; lymphocyte percentages: $F= 10.78$; d.f.= 5, 195; p-value< 0.0001; N:L: $F= 2.56$; d.f.= 5, 195; p-value= 0.029; platelets: $F= 2.96$; d.f.= 5, 120; p-value= 0.015; WBC: $F= 2.29$; d.f.= 5, 195; p-value= 0.048). No treatment effects were found for granulocyte counts ($F= 1.63$; d.f.= 5, 195; p-value= 0.154), hematocrit ($F= 1.43$; d.f.= 5, 123; p-value= 0.209), hemoglobin ($F= 0.45$; d.f.= 5, 199; p-value= 0.810), or percent granulocytes ($F= 3.75$; d.f.= 5, 201; p-value= 0.003, Table 4).

Blood plasma chemistry

Aspartate aminotransferase (1; $F= 7.90$; d.f.= 5,91; $p< 0.0001$; Fig. 1), creatine kinase ($F= 4.77$; d.f.= 5,91; $p= 0.0006$; Fig. 3), glucose ($F= 2.35$; d.f.= 5,91; $p= 0.046$; Fig. 4), and total protein ($F= 9.73$; d.f.= 5,91; $p< 0.0001$; Fig. 5) concentrations were different between transported and control gilts at different transport durations. Aspartate aminotransferase concentrations was greater in transported compared with control gilts at 12 and 18 hours. Creatine kinase was greater in transported gilts compared with control gilts at 12 hours. Glucose concentrations were greater in transported gilts compared with control gilts at 6 hours. Total protein concentrations were greater in transported gilts compared with control gilts at 6, 12, 18, 24, and 30 hours.

Cortisol

Cortisol concentrations were greater in transported gilts compared with non-transported controls at 6 and 24 hours ($F= 3.16$; d.f.= 5,105; p-value= 0.016, Fig. 2).

Cytokines

Interluekin-8 was decreased in transported compared with non-transported control gilts at 6 hours ($F= 6.64$, d.f.= 1,38; $p= 0.014$; Table 2). No other treatment effects were found for the other cytokines measured in this study.

Reproductive performance

There was no transport effects found for any of the reproductive performance variables measured in this study (Table 3). Farrowing rate (Chi-Square= 6.870; d.f.= 5; p-value= 0.231), total born ($F= 1.09$; d.f.= 5, 86; p-value= 0.370), born alive ($F= 0.96$; d.f.= 5, 86; p-value= 0.444), stillborn ($F= 0.88$; d.f.= 5, 86; p-value= 0.497), processing weight ($F= 1.34$; d.f.= 5, 86; p-value= 0.257), number weaned ($F= 0.57$; d.f.= 5, 85; p-value= 0.719), and weaning weight ($F= 1.83$; d.f.= 5, 52; p-value= 0.123).

EXPERIMENT 2 (Fall)

Whole blood hematology

Percent granulocytes, hematocrit levels, lymphocyte counts, N:L ratio, and platelet counts were different ($p<0.05$) among treatment groups (Table 7). (Percent granulocytes: $F= 2.08$; d.f.= 10, 177; p-value= 0.028; Hematocrit: $F= 4.63$; d.f.= 10, 136; p-value< 0.0001; lymphocytes counts: $F= 2.71$; d.f.= 10, 193; p-value= 0.004; N:L: $F= 4.25$; d.f.= 10, 167; p-value< 0.0001; platelets: $F= 5.21$; d.f.= 10,111; p-value< 0.0001).

Granulocyte counts, hemoglobin, and percent lymphocytes did not differ ($p>0.05$) among treatment groups (Table 7). (Granulocytes: $F= 1.43$; d.f.= 10, 188; p-value= 0.168; hemoglobin: $F= 1.46$; d.f.= 10, 167; p-value= 0.157; and percent lymphocytes: $F= 1.47$; d.f.= 10, 183; p-value= 0.155).

Blood plasma chemistry

Albumin, Alk Phos, Bilirubin, BUN, Creat, and GGT were different ($p<0.05$) between transported and non-transported control gilts (Table 5). Aspartate aminotransferase (Fig. 6), CK (Fig. 8), glucose (Fig. 9) and total protein (Fig. 10) were different ($p<0.05$) between transported and non-transported control gilts at different transport durations.

Cortisol

Cortisol concentrations were greater in transported gilts at 12 and 30 hours compared with non-transported controls. ($F= 3.11$; d.f.= 5, 103; p-value= 0.012). Cortisol was significant only when TQA and TQA+ data were combined.

Reproductive performance

There was no transport effects found for any of the reproductive performance variables measured in this study (Table 6). Farrowing rate (Chi-Square= 8.110; d.f.= 5; p-value= 0.150), total born ($F= 1.21$; d.f.= 12, 33; p-value= 0.316), born alive ($X^2= 15.37$; d.f.= 12; p-value= 0.222), stillborn ($X^2= 12.50$; d.f.= 12; p-value= 0.406), processing weight ($F= 1.24$; d.f.= 12, 32; p-value= 0.302), and number weaned ($F= 0.96$; d.f.= 12, 32; p-value= 0.502).

Body weight

There was no difference ($p>0.05$) among initial body weights (Control: 89.5 ± SE1.4 kg; TQA: 91.9 ± SE 1.4 kg; and TQA+ 20%: 92.7 ± SE 1.5kg). Percent weight lost differed among treatments ($F = 66.46$, d.f = 2,104, $P < 0.001$; Figure 11). TQA and TQA+ 20% transported gilts both had greater weight loss than control non-transported gilts. There was no difference in weight lost between TQA and TQA+ 20% transported gilts (multiple comparison $P = 0.658$). The interaction between treatment and time was not significant ($P = 0.072$).

Discussion:

The objective of this study was to assess the effects of long distance transportation on physiology and reproductive performance of breeding-aged gilts. Overall we found little evidence that long distance transport (up to 30 hours) had any major detrimental effects on gilt well-being and reproductive performance.

Cortisol concentrations, the N:L, and glucose concentrations were elevated in transported pigs between 6 to 12 hours compared with control non-transported gilts. This initial increase in these parameters suggest that gilts were experiencing acute stress possibly due to handling, loading onto the trailer, and the early part of transportation. Transportation has been shown previously to increase the N:L ratio and cortisol concentrations in young and adult pigs (McGlone et al., 1983; Sutherland et al., in press). These differences in cortisol, N:L, and glucose concentrations did not persist throughout the 30 hour transport period, suggesting that these animals may have adapting to the stressor of transport.

Dehydration would be expected in transported gilts due to the lack of water during transportation. Both total protein concentrations and body weight change in transported gilts were consistent with mild dehydration. Interestingly, weight loss did not decrease further after 6 hours of transport. McGlone et al. (1993) found a positive relationship between duration of transport and total weight lost, such that pig weights declined throughout the entire trip duration. In the current study, we found weight dropped significantly by 6 hours of transport (~4% total body weight), however, there was little change in weight loss past 6 hours. Gilts transported for 30 hours had the same weight loss as those transported for 6 hours.

As transport duration increased it is likely that pig's experienced increasing amount of hunger and thirst. Lewis et al., (2008) demonstrated that the intestines of market weight pigs were empty after 16 to 18 hours of transport. In the present study, this would have occurred during the 12 to 18 h leg of the trip. As the gut empties during transport gilts would be expected to enter a catabolic state to maintain homeostasis. We found changes in albumin, Alk Phos, AST, bilirubin, BUN, CK, GGT, glucose, and total protein concentrations all of which could indicate a change in metabolism. Creatine kinase concentrations were reported to increase in weaned and market weight pigs during transport (Elbers et al., 1991; Sutherland et al., inpress), probably due to tissue damage caused by muscle exertion as a result of transport. Changes in total protein can reflect dehydration but may also reflect muscle catabolism further supported by the occurrence of elevated BUN (a breakdown product of protein). Sutherland et al. (in press) found that after a 60 minute transport, AST was increased in weaned pigs. Elevated CK and AST concentrations measured in transported pigs was probably due to tissue damage caused by muscle exertion as a result of transport.

Despite the number of physiological changes seen during the acute portion of these studies we found no changes in reproductive performance. The lack of any negative effects of transport on reproductive performance of these sows, suggests that any physiological effects observed in these pigs immediately after transport did not have any long term detrimental effects on gilt reproductive performance. This finding agrees with Hughes et al. (1997) who found that transportation did not affect the onset of puberty in gilts. Friend (2001) reports a similar lack of transportation induced effect on reproduction in horses.

Surprisingly, we found no differences in the physiological response of the gilts to the two different space allowances. Alteration of space allowance may have a more significant effect when applied to an industry setting.

While there were numerous physiological changes seen over the course of the transportation period few were consistent over time or between studies. This lack of consistency raises questions about the biological significance of these results. The lack of a pattern deserves further study as it may be due to effective coping mechanisms or due to the lack of severe consequences due to long distance transportation. In these studies, we followed the TQA guidelines carefully to protect our gilts in this study. Specifically, at every fueling stop during daylight the gilts were sprayed with water (in the summer study). Without this careful management the results may well have been different. Overall, these data indicate that the 28-hour law may be too conservative as we found no overwhelming negative health or well-being effects on breeding gilts after 30 hours of transport as compared to non-transported control gilts.

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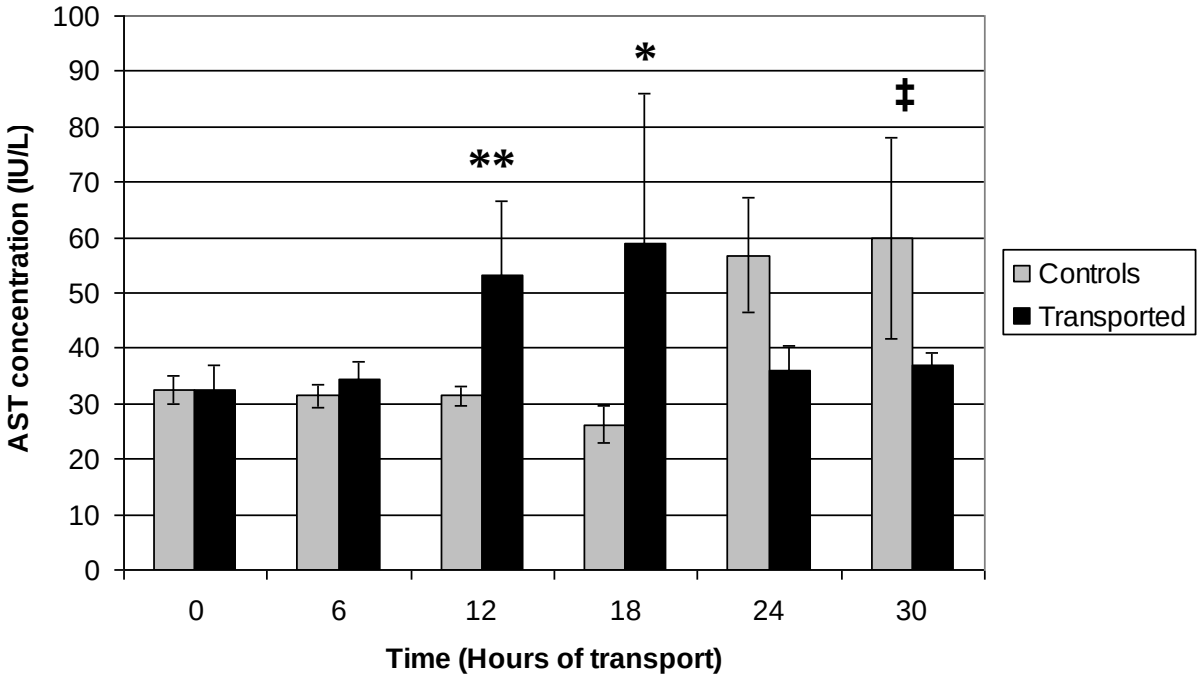

Figure 1. Experiment 1 (Summer) mean (± SE) aspartate aminotransferase (AST) plasma concentrations in transported and control gilts. Transported gilts were transported on a trailer for 6, 12, 18, 24, or 30 hours. Control gilts remained in their home pen with access to food and water. Multiple comparisons are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

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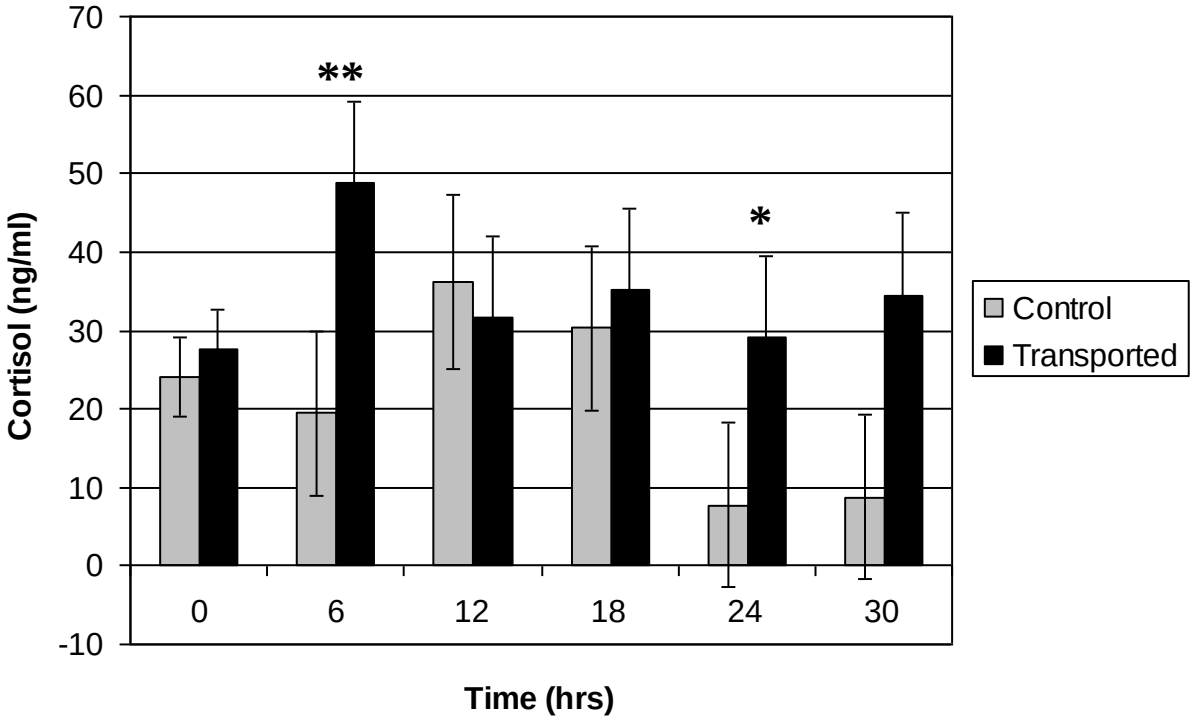

Figure 2. Experiment 1 (Summer) mean (±SE) cortisol concentrations in transported and control gilts. Transported gilts were transported on a trailer for 6, 12, 18, 24, or 30 hours. Control gilts remained in their home pen with access to food and water. Multiple comparisons are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

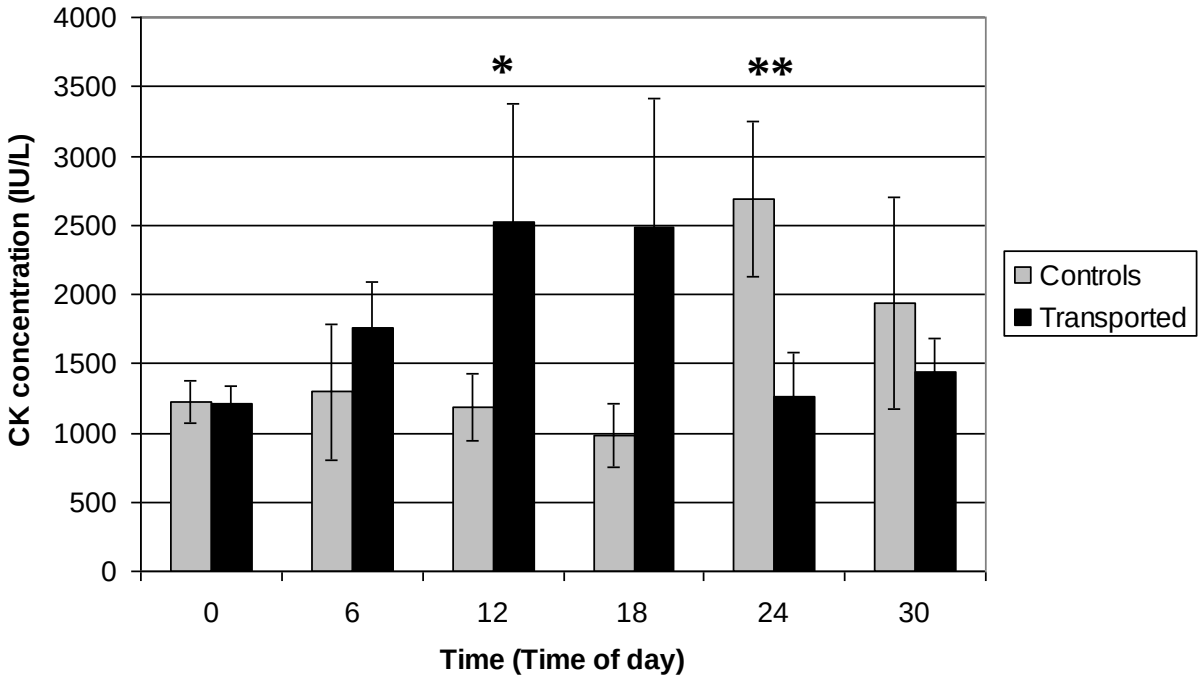

Figure 3. Experiment 1 (Summer) mean (±SE) creatine kinase (CK) concentrations in transported and control gilts. Transported gilts were transported on a trailer for 6, 12, 18, 24, or 30 hours. Control gilts remained in their home pen with access to food and water. Multiple comparisons are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

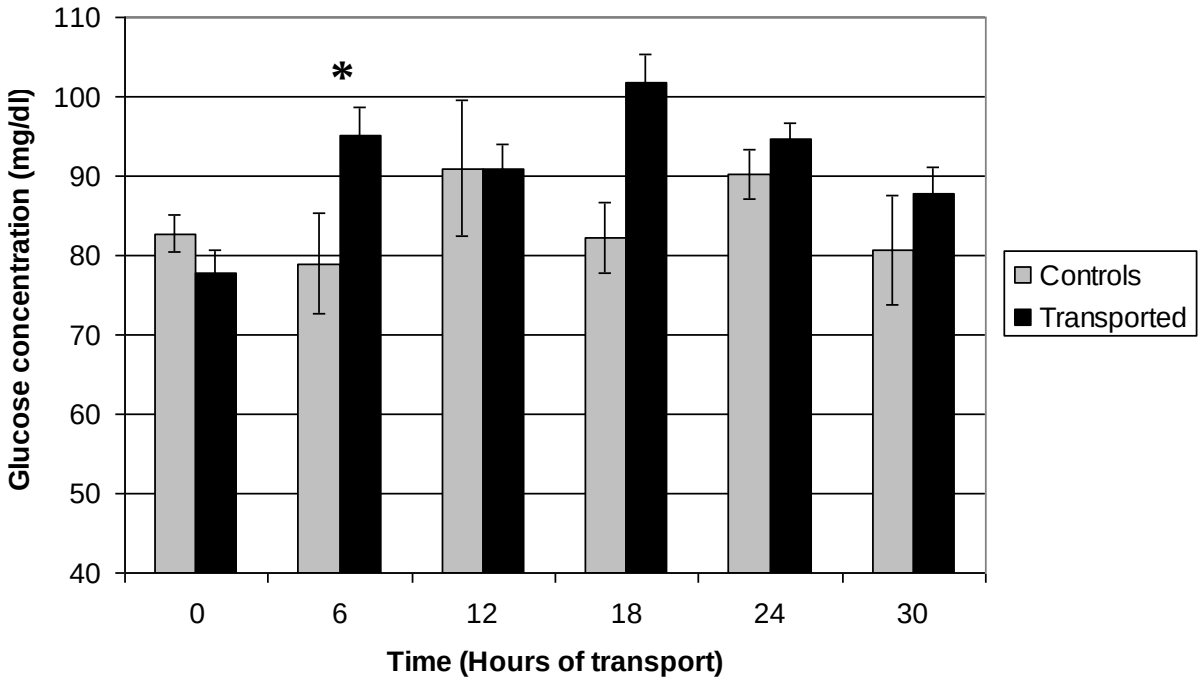

Figure 4. Experiment 1 (Summer) mean (±SE) glucose concentrations in transported and control gilts. Transported gilts were transported on a trailer for 6, 12, 18, 24, or 30 hours. Control gilts remained in their home pen with access to food and water. Multiple comparisons are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

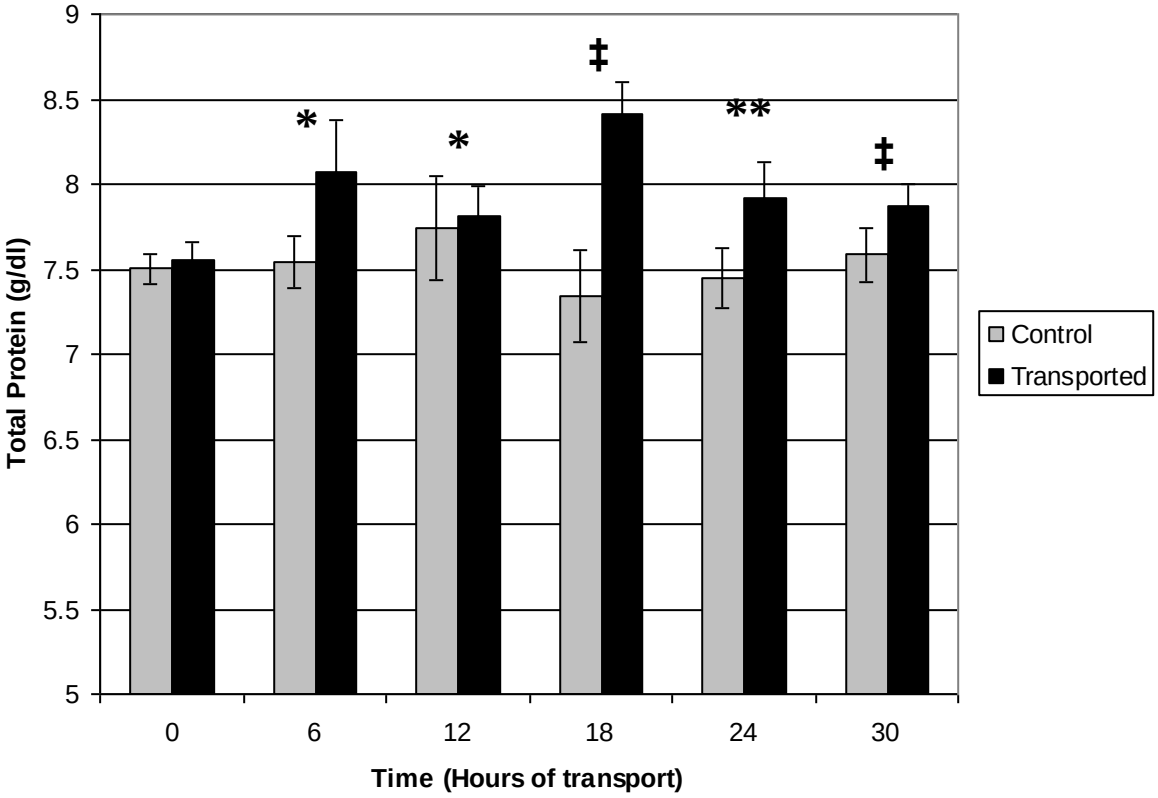

Figure 5. Experiment 1 (Summer) mean (±SE) plasma total protein concentration in transported and control gilts. Transported gilts were transported on a trailer for 6, 12, 18, 24, or 30 hours. Control gilts remained in their home pen with access to food and water. Multiple comparisons are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

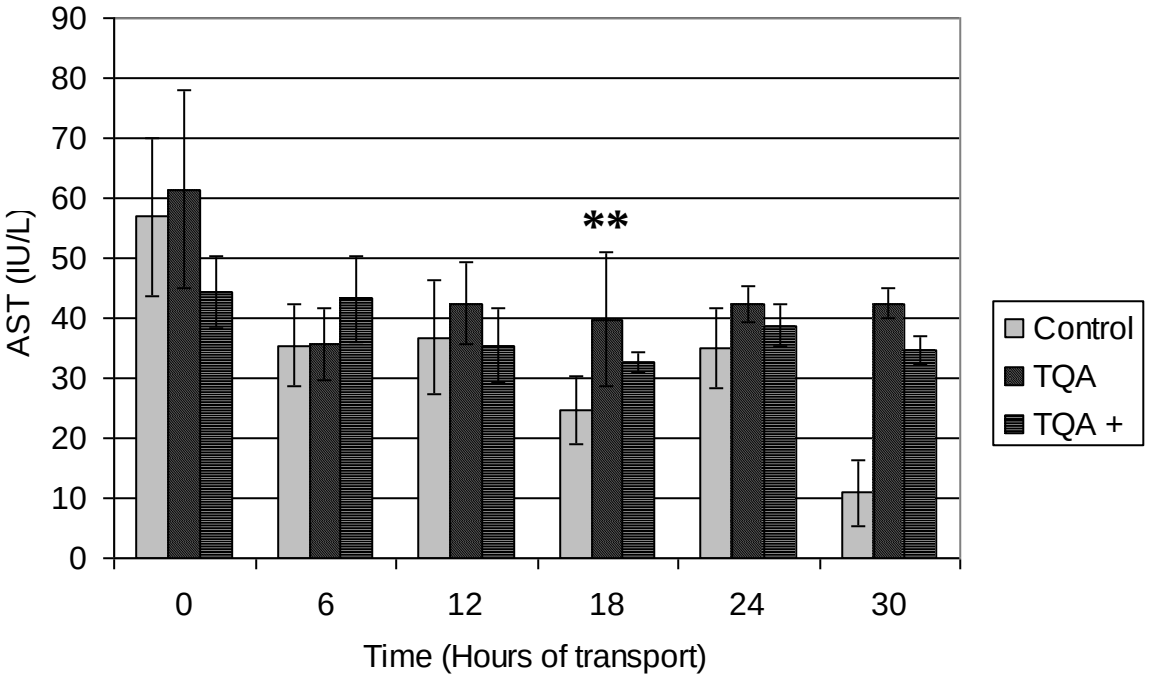

Figure 6. Experiment 2 (Fall) mean (±SE) plasma AST concentrations in gilts. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. [1] Control gilts remained in their home pen with access to food and water. The analysis is significant when TQA and TQA+ are combined. Multiple comparisons denote comparisons between controls and transported and are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

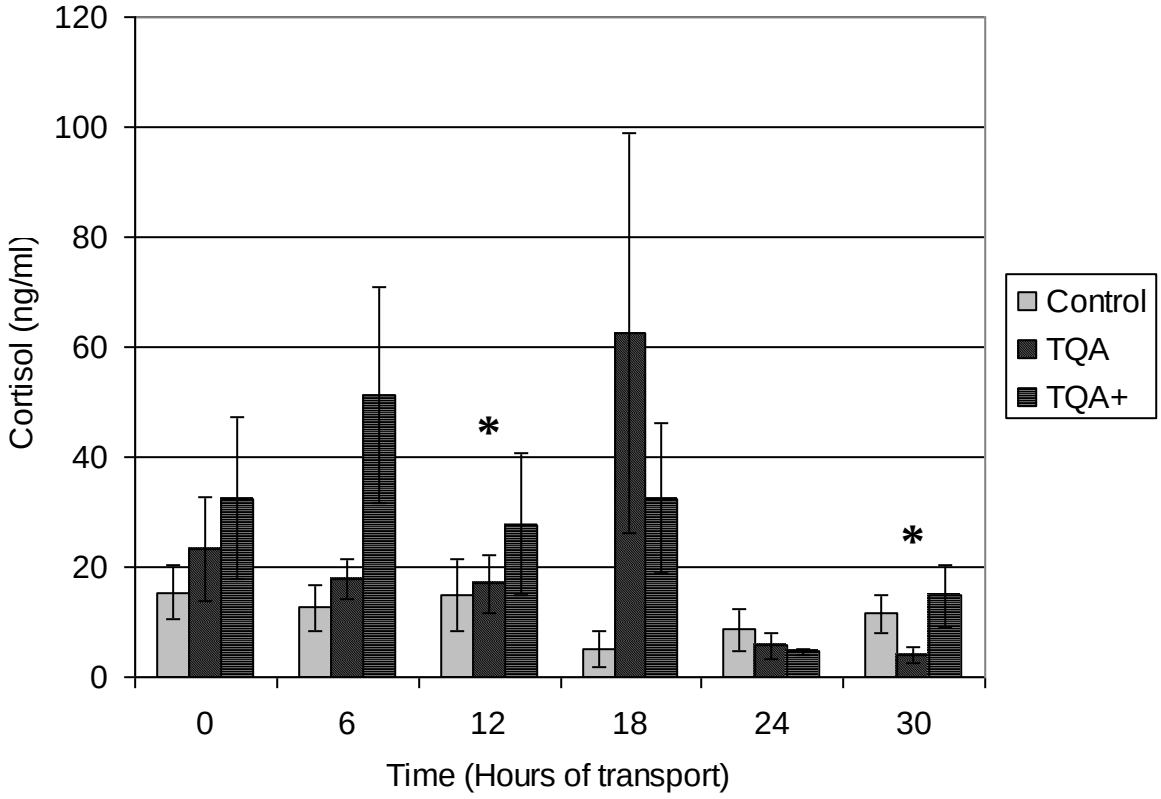

Figure 7. Experiment 2 (Fall) mean (±SE) plasma cortisol concentrations in gilts. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. [1] Control gilts remained in their home pen with access to food and water. The analysis is significant when TQA and TQA+ are combined. Multiple comparisons denote comparisons between controls and transported and are marked when significantly different as follows: '*' for $p < 0.05$; '**' for $p < 0.01$; '‡' for $p < 0.0001$.

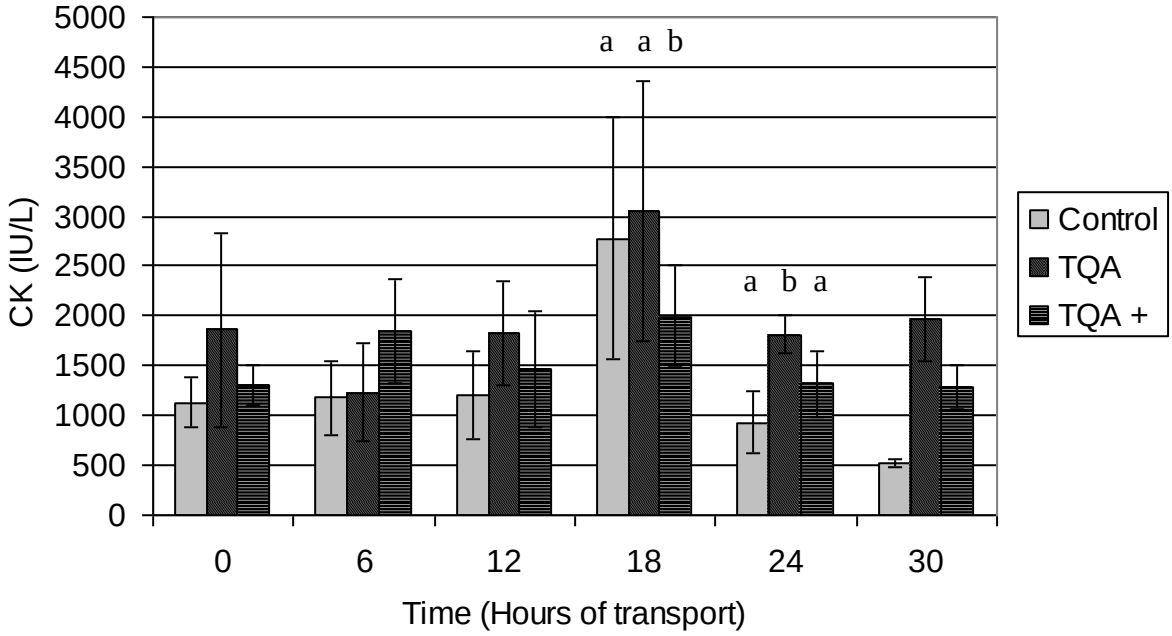

Figure 8. Experiment 2 (Fall) mean (±SE) plasma CK concentrations in gilts. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. Control gilts remained in their home pen with access to food and water. Multiple comparisons denote differences between control and treatment groups (TQA and TQA+) and are reflected in differences in letter.

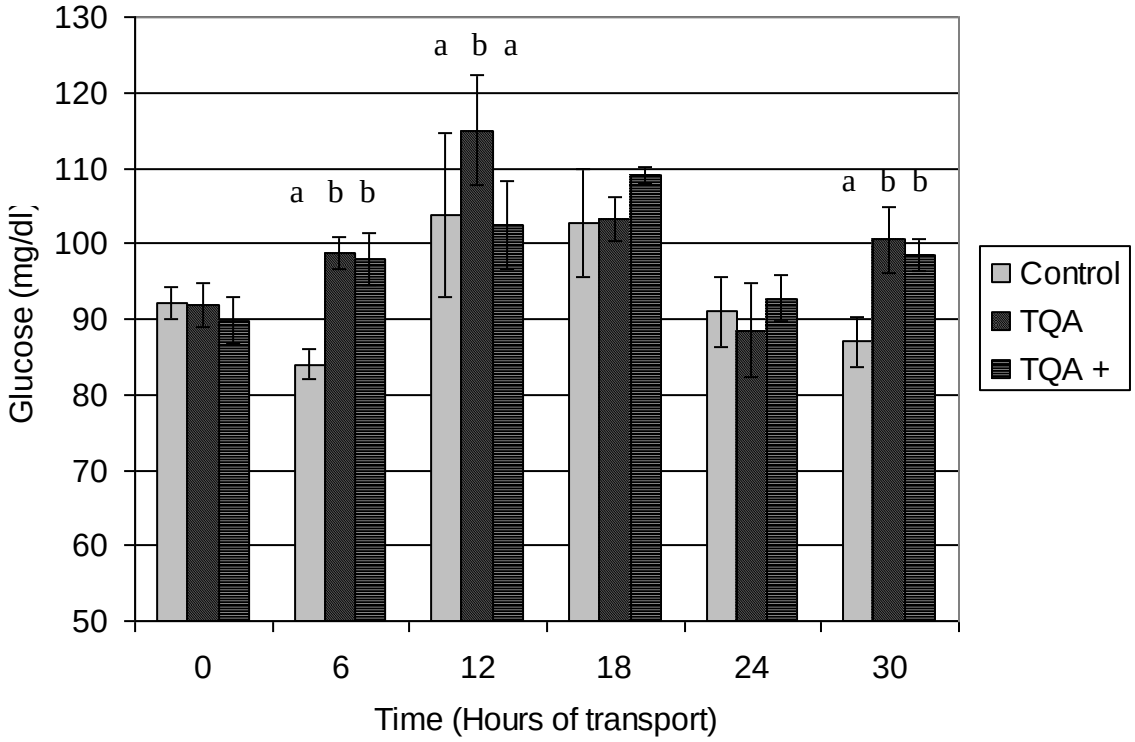

Figure 9. Experiment 2 (Fall) mean (±SE) plasma glucose concentrations in gilts. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. Control gilts remained in their home pen with access to food and water. Multiple comparisons denote differences between control and treatment groups (TQA and TQA+) and are reflected in differences in letter.

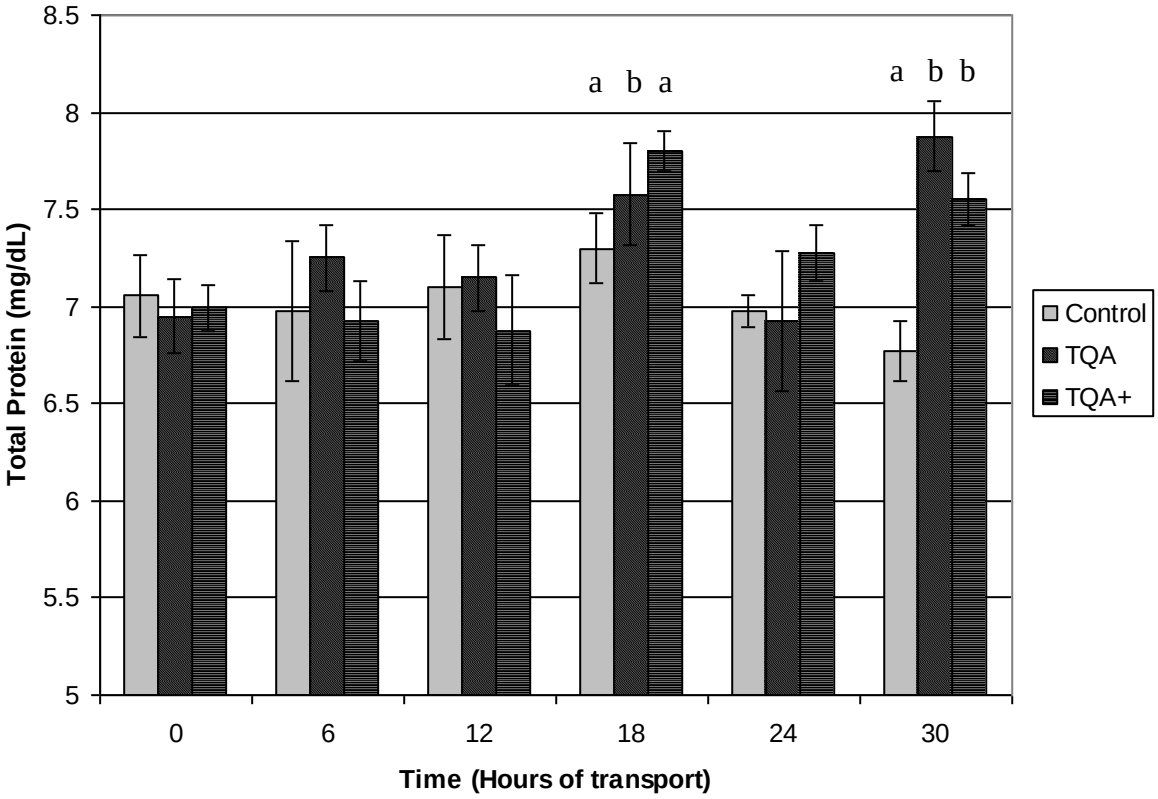

Figure 10. Experiment 2 (Fall) mean (±SE) plasma total protein concentrations in gilts. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. [1] Control gilts remained in their home pen with access to food and water. Multiple comparisons denote differences between control and treatment groups (TQA and TQA+) and are reflected in differences in letter.

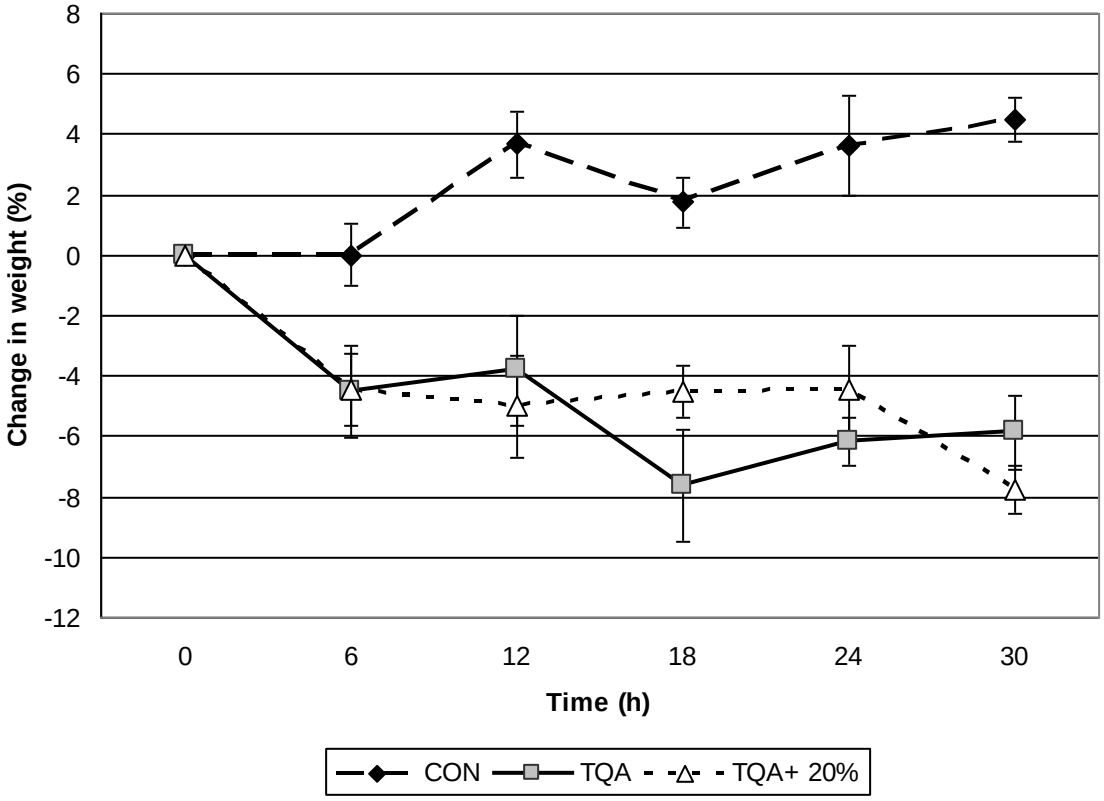

Figure 11. Experiment 2 (Fall) results from weight loss analysis. Gilts were transported on a semi-trailer for various durations (6-30h) in two space allowances. [1] Control gilts remained in their home pen with access to food and water. Controls differed from transported gilts (P< 0.001). There were no differences between space allowances (P= 0.658). There was no time by treatment interaction (P= 0.072).

[1]TQA allowed 0.334 m^2/pig; TQA+ 20% allowed 0.409 m^2/pig.

Table 1. Experiment 1 (Summer) mean (±SE) blood plasma chemistry values of gilts transported from 0 to 30 hours.[1,2]

	Time 0		6 Hrs		12 Hrs		18 Hrs		24 Hrs		30 Hrs	
	Con	Trans	Con	Trans	Con	Trans	Con	Trans	Con	Trans	Con	Trans
Albumin	4.20	4.37	4.26	4.633	4.04	4.65	**4.26**	**4.867**	4.433	4.386	**4.233**	**4.714**
(±SE)	0.077	0.047	0.117	0.174	0.284	0.148	0.117	0.12	0.049	0.026	0.128	0.034
Alk Phos	105.80	101.62	123	100.8	101.2	117.2	106.2	94.83	113.2	98.43	100.3	88
(±SE)	8.257	6.742	28.99	16.77	15.03	24.32	11.43	7.786	19.43	7.852	17.79	5.765
Bilirubin	0.09	0.10	**0.098**	**0.088**	**0.098**	**0.15**	0.102	0.182	0.112	0.126	0.147	0.147
(±SE)	0.007	0.005	0.009	0.024	0.01	0.012	0.017	0.028	0.024	0.007	0.046	0.022
BUN	12.28	13.78	13.88	16.05	12.04	14.55	**14.16**	**19.2**	11.12	13.71	14.18	14.06
(±SE)	0.625	0.535	0.351	1.902	0.507	0.501	2.375	2.537	0.559	1.048	1.369	0.788
Creatinine	1.89	1.89	2.04	2.02	1.90	1.86	1.76	1.82	2.05	1.86	2.38	2.15
(±SE)	0.1	0.6	0.1	0.2	0.2	0.1	0.2	0.12	0.1	0.1	0.33	0.1
GGT	59.24	64.59	51	46.67	44.2	54.83	54.8	60	48.33	51.43	58.17	60.29
(±SE)	9.590	5.883	11.49	7.214	5.21	10.13	8.663	9.504	14.3	10.68	20.92	10.78

[1] Alk Phos, alkiline phosphatase; BUN, blood urea nitrogen; GGT, gamma glutyltransferase

[2] Bold indicates significant ($p < 0.05$) multiple comparisons between control and transported gilts at the specified timepoint.

Table 2. Experiment 1 (Summer) mean cytokine response of gilts transported for 6 and 30 hours.[1]

	6 Hours		30 Hours		Model statistics		
	Control	Transported	Control	Transported	F	d.f.	p-value
Interluekin-1b	787.1	336.4	844.4	527.6	0.07	1, 38	0.800
(±SE)	293.6	80.1	474.7	147.0			
Interluekin-2	102.3	269.5	115.9	371.6	0	1, 36	0.945
(±SE)	43.3	120.4	27.0	92.3			
Interluekin-4	93.8	139.5	203.3	220.4	0.1	1, 36	0.751
(±SE)	22.8	50.8	113.8	54.6			
Interluekin-6	590.0	686.4	420.0	1343.5	0.8	1, 35	0.376
(±SE)	425.2	321.8	95.1	439.1			
Interluekin-8	**317.5**	**73.9**	73.1	129.4	6.64	1, 38	0.014
(±SE)	147.5	21.7	25.1	47.1			
Interluekin-10	1699.5	945.8	1241.9	1030.7	0.29	1, 38	0.592
(±SE)	681.7	296.9	872.0	260.5			
Interluekin-12p-70	9.0	20.2	6.3	28.8	0.13	1, 37	0.717
(±SE)	4.9	9.1	1.5	8.1			
Gamma Interferon	5705.3	1760.2	2328.0	2238.2	3.65	1, 38	0.064
(±SE)	2098.3	487.6	1654.9	617.3			

[1] Bold indicates significant ($p < 0.05$) multiple comparisons between control and transported gilts at the specified timepoint.

Table 3. Experiment 1 (Summer) mean reproductive values of gilts transported from 0 to 30 hours. No significant differences were found.

	Control	6 Hrs	12 Hrs	18 Hrs	24 Hrs	30 Hrs
Total Born	9.79	10.90	9.56	8.45	9.70	10.76
(±SE)	0.52	0.95	0.88	1.04	0.84	0.63
Born Alive	8.75	9.50	8.38	7.91	9.00	10.12
(±SE)	0.60	0.75	0.94	0.95	0.80	0.61
Stillborn	0.86	1.40	1.19	0.55	0.70	0.65
(±SE)	0.27	0.34	0.36	0.31	0.34	0.24
Processing Weight	3.79	4.77	3.93	3.76	4.05	4.54
(±SE)	0.27	0.24	0.45	0.55	0.20	0.21
Number Weaned	7.14	8.20	7.50	7.20	7.70	8.53
(±SE)	0.68	0.57	0.84	1.00	0.76	0.53
Weaning Weight	12.80	14.55	14.16	14.07	14.27	15.04
(±SE)	0.57	0.55	0.53	1.13	0.45	0.75

Table 4. Experiment 1 (Summer) whole blood hematology of gilts transported for various durations.[1]

	Time 0		6 Hrs		12 Hrs		18 Hrs		24 Hrs		30 Hrs	
	Con	Trans	Con	Trans	Con	Trans	Con	Trans	Con	Trans	Con	Trans
Granulocytes	4.65	4.44	4.32	4.23	4.68	4.20	4.18	4.46	4.88	4.89	4.86	4.48
(±SE)	0.292	0.184	0.360	0.557	0.968	0.369	0.717	0.362	0.492	0.446	0.667	0.313
% Granulocyte	22.3	22.2	23.1	21.3	**25.0**	**22.3**	19.5	22.5	22.4	23.6	**21.6**	**21.2**
(±SE)	1.2	0.7	2.8	2.2	3.8	1.6	2.9	1.8	1.7	1.3	2.3	1.5
Hematocrit	0.340	0.333	0.336	0.319	0.326	0.321	0.348	0.344	0.356	0.330	0.337	0.349
(±SE)	0.006	0.007	0.004	0.026	0.015	0.020	0.007	0.008	0.014	0.013	0.019	0.007
Hemoglobin	119.8	117.2	118.4	110.8	115.8	117.3	121.6	120.5	122.8	115.4	120.8	121.8
(±SE)	1.91	2.40	2.68	8.82	6.29	6.25	3.20	2.85	3.07	4.14	5.14	2.11
Lymphocytes	12.00	11.80	**11.22**	**10.20**	**9.94**	**11.22**	**11.74**	**12.01**	12.53	12.19	**14.23**	**13.25**
(±SE)	0.50	0.35	1.05	1.28	1.05	0.93	0.76	0.60	1.27	1.21	0.77	0.74
% Lymphocyte	57.9	59.3	**58.5**	**57.5**	**54.7**	**60.4**	**58.4**	**58.8**	57.5	58.2	**60.0**	**61.7**
(±SE)	1.0	0.8	3.4	2.2	2.3	1.5	2.3	1.6	2.2	1.9	1.4	1.7
MIDS	4.09	3.70	3.60	3.84	3.58	3.48	4.44	3.39	4.30	4.19	4.48	3.64
(±SE)	0.21	0.16	0.78	0.50	0.26	0.30	0.42	0.20	0.36	0.46	0.38	0.27
% Mids	19.8	18.4	18.4	21.2	20.3	17.4	22.1	18.0	20.2	18.4	18.2	17.1
(±SE)	1.0	0.5	3.3	1.6	2.6	1.0	2.5	1.1	2.5	1.1	1.4	0.8
N:L	0.380	0.388	**0.377**	**0.409**	**0.372**	**0.455**	0.377	0.336	0.415	0.390	**0.345**	**0.360**
(±SE)	0.01	0.02	0.04	0.07	0.03	0.07	0.03	0.05	0.03	0.03	0.02	0.04
Platelets	294.2	267.4	**243.8**	**175.5**	290.2	247.6	201.4	310.7	**237.0**	**292.4**	200.8	304.2
(±SE)	32.9	14.5	43.6	37.0	89.1	25.8	54.9	31.1	36.1	31.1	108.5	25.9
WBC	20.18	20.00	**19.16**	**19.16**	**18.22**	**18.76**	20.07	20.05	21.70	20.81	**23.77**	**21.33**
(±SE)	0.919	0.598	1.235	0.900	1.699	0.624	0.998	0.990	1.466	1.881	1.413	0.571

[1]Bold highlighting indicates significant difference ($p<0.05$) between pair of values at the selected time point.

Table 5. Experiment 2 (Fall) mean (±SE) blood plasma chemistry values of gilts transported from 0 to 30 hours at two different space allowances.[1,2]

	Space allowance	Duration of Transport											
		0		6		12		18		24		30	
		Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE
Albumin (g/dL)	CON	3.90	0.2	3.85[a]	0.2	3.80	0.2	4.08	0.2	4.00[a]	0.2	3.70[a]	0.2
	TQA	3.85	0.2	4.03[a]	0.2	4.18	0.2	4.28	0.2	3.78[b]	0.2	4.05[b]	0.2
	TQA+	3.97	0.2	3.78[b]	0.2	4.18	0.2	4.28	0.2	4.08[b]	0.2	4.43[b]	0.2
Alk Phos (IU/L)	CON	115	16	101	14	126	14	106[a]	14	110	14	124[a]	14
	TQA	125	16	99	14	131	14	92[a]	14	102	14	133[b]	14
	TQA+	116	16	120	14	112	14	115[b]	14	116	14	103[a]	14
Bilirubin (mg/dL)	CON	0.075	0.02	0.075	0.02	0.080	0.02	0.055[a]	0.02	0.068	0.02	0.055[a]	0.02
	TQA	0.097	0.02	0.110	0.02	0.085	0.02	0.108[b]	0.02	0.148	0.02	0.103[b]	0.02
	TQA+	0.070	0.02	0.105	0.02	0.080	0.02	0.115[b]	0.02	0.088	0.02	0.103[a]	0.02
BUN (mg/dL)	CON	10.7	1.2	10.5	1.7	13.0	1.7	9.9	1.7	9.9	1.7	12.3[a]	1.7
	TQA	11.0	1.2	10.9	1.7	10.9	1.7	12.1	1.7	13.0	1.7	10.0[b]	1.7
	TQA+	11.9	1.2	12.6	1.7	18.4	1.7	13.3	1.7	12.0	1.7	9.9[b]	1.7
Creatinine (mg/dL)	CON	1.55	0.2	1.38	0.2	1.75	0.2	1.43	0.2	1.55	0.2	1.45	0.2
	TQA	1.81	0.2	1.73	0.2	1.75	0.2	1.50	0.2	2.05	0.2	1.65	0.2
	TQA+	1.82	0.2	1.58	0.2	1.68	0.2	1.53	0.2	1.55	0.2	1.73	0.2
GGT (IU/L)	CON	0.075	0.02	0.075	0.02	0.080	0.02	0.055[a]	0.02	0.068	0.02	0.055[a]	0.02
	TQA	0.097	0.02	0.110	0.02	0.085	0.02	0.108[b]	0.02	0.148	0.02	0.103[b]	0.02
	TQA+	0.070	0.02	0.105	0.02	0.080	0.02	0.115[b]	0.02	0.088	0.02	0.103[a]	0.02

[1] Alk Phos, alkiline phosphatase; BUN, blood urea nitrogen; GGT, gamma glutyltransferase

[2] Superscripts between different letters indicate siginficant differences ($p<0.05$) between control and treatment values at the selected timepoint.

Table 6. Experiment 2 (Fall) mean reproductive values of gilts transported from 0 to 30 hours. No significant patterns were found. No data available for number weaned at 24 hrs.

	Control	6 Hrs	12 Hrs	18 Hrs	24 Hrs	30 Hrs
Total Born	9.2	11.5	11.6	10.4	6.3	12.8
(±SE)	1.1	1.7	1.3	1.1	2.5	1.2
Born Alive	9.0	10.6	10.8	10.4	4.8	10.8
(±SE)	1.0	1.5	1.3	1.0	2.5	1.2
Stillborn	0.2	0.9	0.8	0.0	1.5	2.0
(±SE)	0.2	0.8	0.3	0.1	0.0	0.0
Processing Weight (lbs)	4.6	3.4	4.4	4.2	3.5	3.6
(±SE)	0.7	1.2	0.6	0.3	1.7	0.8
Number Weaned	7.0	9.9	8.4	9.5	-	10.0
(±SE)	1.4	1.6	1.8	0.6	-	1.5

Table 7. Experiment 2 (Fall) mean (±SE) whole blood hematology for gilts transported various durations[1,2]

	Space allowance	Duration of Transport											
		0		6		12		18		24		30	
		Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE	Mean	±SE
Granulocyte	CON	2.39	0.28	4.55	0.60	5.16	0.89	5.25	0.82	4.66	0.71	5.33	0.78
	TQA	2.80	0.34	4.80	0.68	3.94	0.65	5.15	1.25	3.96	0.65	3.76	0.97
	TQA+	2.34	0.29	5.97	0.83	3.22	0.43	4.15	0.85	4.27	0.55	3.71	0.87
%Gran	CON	13.8	10.1	19.9[a]	20.3	23.5	21.7	25.7	28.7	17.9[a]	20.3	55.8[a]	21.7
	TQA	10.3	10.1	28.2[a]	19.2	20.8	21.7	31.2	28.7	21.2[a]	20.3	21.6[b]	21.7
	TQA+	24.8	9.8	34.4[b]	21.7	106.3	20.3	22.4	28.7	56.8[b]	20.3	12.0[a]	17.2
Hematocrit (%)	CON	0.33	0.02	0.31[a]	0.01	0.34[a]	0.01	0.34	0.01	0.27[a]	0.01	0.33	0.01
	TQA	0.32	0.02	0.29[b]	0.01	0.35[a]	0.01	0.34	0.02	0.31[b]	0.01	0.35	0.02
	TQA+	0.34	0.02	0.35[a]	0.02	0.33[b]	0.01	0.37	0.02	0.34[b]	0.01	0.36	0.02
Hemoglobin (g/dL)	CON	93.8	16.3	107.4	12.9	119.5	13.8	95	18.2	86.6	12.9	100.3	14.9
	TQA	90.6	16.1	81.2	13.8	122.7	13.8	118.6	25.6	98.5	12.9	79.2	13.8
	TQA+	96.2	16.4	123.4	14.9	98	14.9	73.6	18.3	116.6	13.8	65.2	14
Lymphocytes	CON	15.14	0.94	12.41[a]	1.19	11.59	1.57	11.28	1.88	14.97[a]	2.97	13.44[a]	1.79
	TQA	15.24	1.75	7.66[b]	1.01	10.43	1.22	7.00	0.62	9.59[a]	1.09	8.90[b]	1.23
	TQA+	12.54	0.88	7.13[b]	0.62	9.22	1.44	10.13	0.54	9.84[b]	1.25	12.60[a]	0.85
%Lym	CON	59.2	4.4	55.4	4.4	49.1	4.4	54.2	6.3	61.5	4.4	47.3	4.7
	TQA	62.6	4.4	48.8	4.2	52.1	4.4	45.3	6.3	53.7	4.4	52.9	4.4
	TQA+	57.2	4.3	43.3	4.7	48.2	4.4	56.6	6.3	46.5	4.4	56.4	4.4
MIDS	CON	6.33	0.34	5.85	1.06	5.17	0.79	4.08	0.50	4.70[a]	0.46	5.81[a]	0.79
	TQA	6.22	0.41	3.83	0.53	4.29	0.42	3.70	0.43	4.48[a]	0.55	3.49[b]	0.78
	TQA+	5.73	0.62	4.10	0.58	3.20	0.53	3.73	0.26	4.96[b]	0.49	4.17[b]	0.36
%Mids	CON	37.3	27.3	24.8	30.7	51.4	30.7	20.2	43.5	20.6[a]	30.7	69.7	32.9

	TQA	32.2	26.9	23.0	29.0	54.8	30.7	23.6	43.5	25.1[a]	30.7	66.0	30.7
	TQA+	58.3	26.4	23.1	32.9	104.0	30.7	21.0	43.5	67.5[b]	30.7	66.2	29.6
N:L	CON	0.20	0.0	0.37[a]	0.1	0.45	0.1	0.48	0.1	0.30[a]	0.1	0.49	0.1
	TQA	0.17	0.0	0.62[b]	0.1	0.37	0.1	0.72	0.1	0.40[a]	0.1	0.40	0.1
	TQA+	0.22	0.0	0.85[b]	0.1	0.42	0.1	0.40	0.1	0.48[b]	0.1	0.24	0.1
Platelets	CON	218.4	49	342[a]	38	212.1[a]	39.4	198[a]	62.2	232.7[a]	40.7	233.2[a]	45
(K/µL)	TQA	184.2	48.2	215.4[b]	44.4	276.7[b]	40.7	245.9[b]	65.6	301.6[b]	40.7	376.9[b]	49.7
	TQA+	254.6	48.9	317.8[a]	41.1	197.6[a]	44.8	432.3[b]	69.3	282.1[a]	40.7	333.5[b]	53.6
WBC	CON	22.4	3.2	22.9	2.3	20.0	2.3	20.6	3.3	22.1	2.3	23.2	2.5
(K/µL)	TQA	20.9	3.1	16.2	2.2	15.9	2.3	15.9	3.3	18.0	2.3	14.9	2.3
	TQA+	22.6	3.1	17.3	2.5	14.8	2.3	18.0	3.3	18.8	2.3	19.4	2.4

[1]Superscripts indicate siginficant differences between different letters within space allowance by time ($P < 0.05$)
[2]WBC, white blood cell; %Lym, percent lymphocyte; %Mids, includes monocytes, eosinophils, basophils, promonocytes, blasts; %Gran, percent granulocytes includes segmented and banded neutrophils, metamyelocytes, myelocytes, promeyelocytes; N:L percent graulocyte to percent lymphocyte ratio