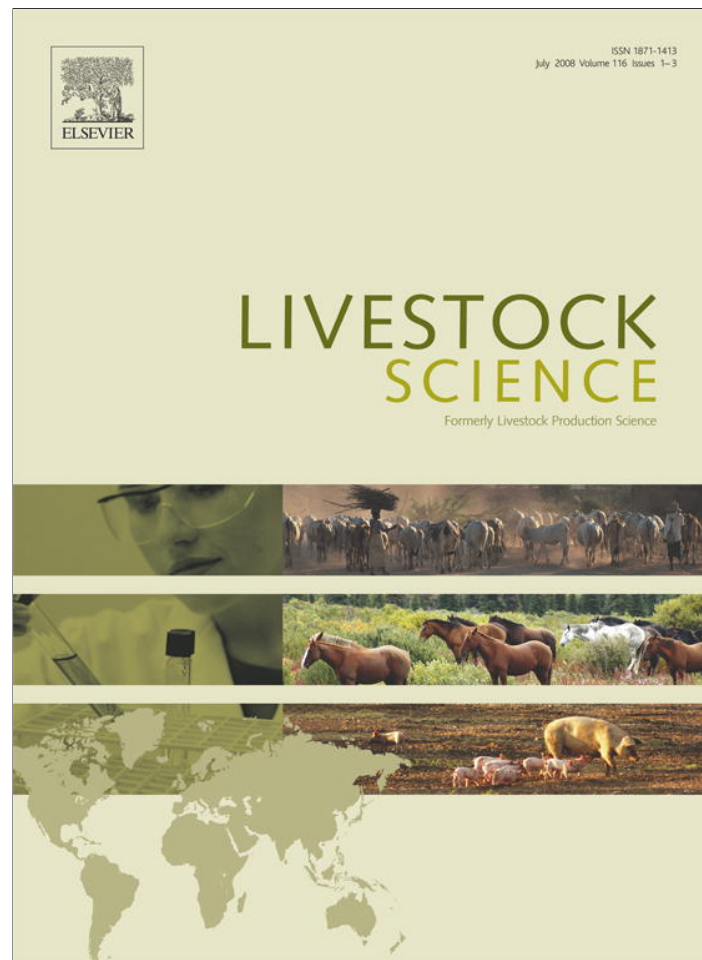


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Short communication

Novelty causes elevated heart rate and immune changes in pigs exposed to handling, alleys, and ramps

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Abstract

The objective of the present research was to quantify the cardiovascular and physiological responses in pigs subjected to novel alleys and ramps. Ten test subject pigs were selected per treatment. The first (trained) group were trained to navigate a course including a ramp. Testing was daily for seven days. Once training was completed, the trained and control (naïve) groups were exposed to a fixed course, the course and ramp (both up and down) while heart rates, time, handling difficulty, and blood were collected to determine the innate responses. Heart rates of trained pigs were reduced significantly ($P=0.003$) compared to naïve pigs travelling the same course. Both handling ease and handling time were significantly improved for the trained pigs ($P=0.03$ and $P=0.01$ respectively) compared to naïve pigs. Blood immune measures indicated reduced stress among trained pigs who had lower neutrophil numbers ($P=0.04$) and lower total and average phagocytosis ($P=0.001$ and $P=0.02$) compared with naïve pigs. This study demonstrated that the exposure of pigs to a novel environment clearly causes a mild physiological response. Pigs are not inherently stressed by alleys and ramps, but rather novel experiences cause handling problems and a stress response and minimal training can reduce the stress experience for the pig.

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Keywords: Pig; Handling; Novel environment; Stress**1. Introduction**

Economic losses related to the handling of pigs are well known throughout the industry (Tarrant, 1989). Handling effects on pig stress are complicated and few studies have quantified stress responses in common handling situations.

In times of stress, the HPA (hypothalamic pituitary-adrena axis) axis is activated and glucocorticoids are released into the blood of the pig. Stress also causes changes in measures of the immune system (McGlone et al., 1993; Morrow-Tesch et al., 1994; Hicks et al., 1998; Salak-Johnson and McGlone, 2007). Alongside endocrine and immune responses to stress, stressed animals may undergo physiological changes such as increased heart and respiration rates that lead to an increase in body core temperature. Heart rate can be determined by use of telemetry to obtain undisturbed responses within experimental groups (Von Borell, 2001; Von Borell et al., 2007; Marchant-Forde et al., 2003a,b).

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The main hypothesis for this study was that when pigs are exposed to a novel environment they would have a higher heart rate and an overall stress response compared with those pigs that were habituated with the environment. We also sought to determine if minimal training to remove novelty would reduce the stress experience for pigs destined for transport to slaughter. The ultimate goal is to reduce or eliminate stress-induced problems with pig losses during transportation and handling.

2. Materials and methods

This experiment was approved by the Texas Tech University Animal Care and Use Committee. The subjects chosen for experimentation were progeny of the Camborough 22 (C-22), PIC USA breeding stock located at the Texas Tech University swine unit. Pigs were randomly selected within established penned groups from a standard indoor finishing barn and assigned to a treatment, and they remained in the pens for the duration of the experiment. For inclusion within this study the individual pigs needed to be at or over 90 kg live weight and not treated with medications or feed additives. All pigs were naïve to handling in the manner of this study.

The novel environment for all test pigs was a course and a ramp. The ramp was custom built for this project and had a 35° angle to the peak (which is a similar angle to many ramps within livestock trailers), with 4 ft long sides and had cleats (small steps spaced inches apart) to aid the pig during ascent and descent. The ramp was also built wide enough so that more than three pigs could cross the ramp at one time. The ramp was placed in a standard handling course that was constructed within the finishing barn that consisted of two straight hallways, five turns, and movement through a confined area. The handling course is similar to the one described in detail by McGlone et al. (2004) but with a ramp added. Ten pigs were exposed to the ramp once a day for seven days. After the seven days of training, all the animals (trained and naïve) were run through the model on the eighth day in two groups of three and one of four per treatment.

Cardiovascular data were collected using the Polar Sports 610 ir heart rate monitor and T61 coded transmitter (Polar Electro, Oy, Finland). This heart rate monitor was attached to three of the pigs within an individual group so each treatment group had data from nine animals overall. The device consists of an electrode belt with a transmitter and a watch receiver. The receiver stored the data transmitted from the belt. The receiver was set to receive heart rate data every 5 s. The belt was fit around the pig's chest, using a canvas and velcro constructed belt strap with a pouch to contain the monitor (Southwest Canvas, Lubbock, TX). A lubricant gel was applied to the sensors so that a good connection was maintained with the pig's skin. During collection of data, pigs were gently prompted away from other curious pigs to prevent other pigs from removing the belt or receiver or otherwise interfering with the device. However, some pig-device interference resulted in lost information.

After collection of a full data set, the belt and receiver were removed from the pigs, and the receivers were taken to a computer outfitted with the Polar downloading receiver. Data were then downloaded.

Data were grouped into a mean heart rate for pre-handling, during exposure to the ramp, and post-exposure periods. Other handling-associated measures taken from the experiment were time to navigate the ramp, and a subjective handling score. The handler based the score on a four point scale where one is minimal difficulty to move and four is very difficult to move requiring active the use of a livestock board.

Blood was drawn over lithium heparin immediately after experimentation so that immune measures associated with stress could be analyzed. The pigs were restrained using a nose loop via a process previously established as minimally stressful to the pig (Lewis et al., 2005). The blood was collected from the jugular vein using heparinized tubes, spun and separated, and analyzed using the Cell Dyne instrument (Abbott Labs, Santa Clara, CA) to count total leukocytes and determine leukocyte differentials. The plasma was then frozen until cortisol was measured using a Coat-a-Count cortisol kit (Diagnostic products, Los Angeles, CA) following the manufacturer's protocol.

Neutrophil chemotaxis, chemokinesis, and phagocytosis were determined on whole blood by methods previously described by McGlone et al. (1993) and Hulbert and McGlone (2006). Briefly, for chemotaxis and chemokinesis a modified Boyden chamber (Neuro Probe, Cabin John, MD) was used to measure the migration of neutrophils toward media (chemokinesis) or toward 10^{-8} M of recombinant human complement fragment, C5a, (chemotaxis). The number of beads phagocytized was counted as 0, 1, 2, 3, 4, 5 or 6 or more. The percentage of cells that phagocytized at least one bead and the average numbers of beads phagocytized were determined.

Table 1
Effects of training or naivety on pig physiological measures when exposed to a novel handling facility

Measure	Trained	Naïve	SE	<i>P</i> -value
Heart rate (bpm)	151.2	172.6	4.16	0.003
Handling score (1–5 scale)	1.33	3.33	0.32	0.01
Time to complete course (min)	1.26	3.97	0.63	0.03
Weight of pigs (lb)	221.3	217.0	9.20	0.75
White blood cells (no/mL × 10 ⁻³)	12.28	18.63	1.94	0.08
Neutrophils (no/mL × 10 ⁻³)	3.08	4.48	0.34	0.04
Lymphocytes (no/mL × 10 ⁻³)	7.75	12.3	1.60	0.11
Monocytes (no/mL × 10 ⁻³)	0.66	1.00	0.11	0.11
Eosinophils (no/mL × 10 ⁻³)	0.64	0.72	0.11	0.60
Basophils (no/mL × 10 ⁻³)	0.15	0.15	0.03	0.91
Neutrophil:lymphocyte ratio	0.48	0.40	0.07	0.46
Chemokinesis (no. cells 5 fields/well)	173.6	144.5	25.7	0.47
Chemotaxis (no. cells 5 fields/well)	242.4	306.1	52.5	0.44
Phagocytosis (% cells)	40.4	58.8	1.79	0.001
Average Phagocytosis rate (no. beads engulfed/100 cells)	4.46	7.24	0.55	0.02

Data were analyzed as a completely random design using SAS General Linear Models. The effects in the model included fixed effect of treatment (exposed vs. naïve) and period (pre, during, post-exposure) and the interaction of treatment and period.

3. Results

Trained pigs had a mean handling score of 1.33, while the naïve pigs had a higher score of 3.33 ($P=0.01$; Table 1). Trained pigs required a mean of 1.26 min to complete the course, while the naïve pigs required 3.97 min on average ($P=0.03$) to complete the course.

Heart rates of trained pigs were lower ($P=0.003$) than those of naïve pigs (Table 1, Fig. 1). Plasma neutrophil numbers were higher ($P=0.04$) among naïve pigs compared to control pigs. Both the percentage of neutrophils that phagocytised beads ($P=0.001$) and average number of beads phagocytised ($P=0.02$) were elevated among naïve pigs compared with trained pigs.

Cortisol concentrations did not statistically differ between treatment groups ($P=0.56$). The trained group had 31.0 ng/mL and the naïve group yielded 27.0 ng/mL (SE=4.96).

4. Discussion

Heart rate data demonstrated that pre-conditioning pigs to handling resulted in lower heart rate compared to naïve pigs that experienced the same course. This finding could be due to either pigs (1) were more physically conditioned to the task and thus showed a lower cardiovascular response or (2) they experienced less stress because they were familiar with the course, thus reducing fear of the unknown or finally, (3) they experienced less physical strain due to less handling and quicker solving of that task that has been facilitated through learning from pre-exposure. Pigs are unlikely to

be more fit or conditioned by only 7 days of exercise. It remains to be determined what the minimum amount of pre-exposure to novel settings is required to reduce the stress response of novel handling situations. A future study could try to examine this.

The immune measures supported the cardiovascular results. Morrow-Tesch et al. (1994) found that naïve pigs showed greater stress-induced changes in immune measures. Elevation in the numbers of blood neutrophils and an increase in phagocytosis may be indicators of the stressfulness of the handling experience (Salak-Johnson and McGlone, 2007).

Trained pigs compared with naïve pigs, had lower heart rates, and lower (easier) handling scores (Table 1). Because the plasma cortisol concentrations did not differ between trained and naïve pigs, the stress levels experienced by novel exposure to this course was not great. The mild physical exertion as indicated by reduced cardiovascular activity resulted in a small, acute stress response that was minimized by previous handling or training.

This study used one genetic line of pigs; however it could be postulated that different breeds of pigs would also react differently to a novel environment. For example, Desautels et al. (1999) showed that Large White pigs had greater post-stress levels of adrenocorticotropin hormone (ACTH) than a Meishan line and Sutherland et al. (2005) highlighted a breed effect on baseline immune, cortisol, and performance data. The genetic differences between breeds could have a huge effect on the “stress tolerance” to a pig when confronted by a novel environment. A quantitative genetic study searching for loci associated with stress traits (QTL) could highlight genetic breed differences. Desautels et al. (2002) found a QTL of major effect for basal cortisol levels and also post-stress cortisol levels on *Sus scrofa* chromosome 7. Interestingly it is the Meishan alleles that are associated with the raised cortisol levels and that are partially dominant for the post-stress levels. Results of this type could be used in future breeding programs as part of a breeding index for stress-robustness.

The pre-conditioning of the pigs may reduce the incidence of cardiovascular stress in pigs prior to processing, via exposure to ramps, noise, etc. This exposure may then reduce the incidence of stress involved with handling and transport. If pigs are moved in the optimal group size of 5 or 6 pigs (Lewis and McGlone, 2007), handling may have a less negative effect on meat quality and fatigued pig numbers (Tarrant, 1989).

This study demonstrated that pigs that have been pre-exposed to an environment have lower heart rates, less elevated neutrophil phagocytosis, and handled easier

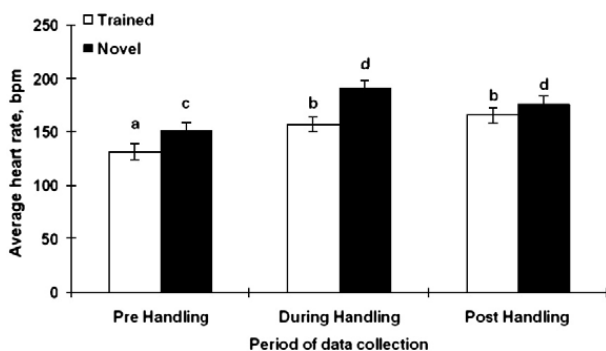


Fig. 1. Mean period differences in heart rate data between treatments at distinct periods during experimentation (letter differences denote a significant difference $P<0.05$) in training within a period.

and in less time than pigs that have had no previous handling experience. Therefore, the potential dilemma of handling pigs from farm to processing plant is not due to the nature of a familiar handling facility (ramps up, down, turns, etc.) but rather the novelty of the environments that pigs may experience.

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