Core body temperature does not cool down with skin surface temperature during recovery at room temperature after acute heat stress exposure

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ABSTRACT

Identifying new methods of assessing livestock welfare is a growing area of research. Non-invasive methods such as infrared thermography are valuable for quick and accurate observations and could be utilized to monitor the thermal status of swine without direct contact. The study objective was to determine if infrared thermography could be used as a non-invasive, hands-off approach to accurately monitor the welfare of swine by comparing changes in skin surface temperature (SST), core body temperature (CBT), behavior, and heart rate during acute heat stress (AHS) and subsequent recovery in thermoneutral (TN) conditions. In eight replications, 16 pigs ($n = 8$ barrows and 8 gilts) were subjected to AHS ($39.3 \pm 0.1^{\circ}\text{C}$) for 30 min followed by TN ($20.6 \pm 0.1^{\circ}\text{C}$) for 30 min. The SST, CBT, heart rate, and behavioral data were recorded throughout the entire experiment. During pre-treatment, ear base SST was greater ($P < 0.01$; $35.6 \pm 0.3^{\circ}\text{C}$) than all other locations. The SST at all locations increased ($P < 0.01$) linearly with duration of AHS exposure. During TN, maximum CBT was greater ($P < 0.01$; $40.6 \pm 0.1^{\circ}\text{C}$) compared to during AHS ($40.3 \pm 0.1^{\circ}\text{C}$). Pigs spent more time standing during AHS ($P < 0.01$) and tended ($P = 0.10$) to lie more during TN; however, heart rate (141 ± 2.3 beats per minute) was not affected by treatment or duration. In summary, rapid TN exposure after an AHS challenge reduces SST; however, CBT was actually increased and this may have implications towards reduced activity and increased organ damage.

Keywords: heat stress, recovery, infrared thermography, pig, swine

1. Introduction

Commercially raised pigs are subjected to multiple environmental stressors throughout their lifetime. Due to a lack of functional sweat glands (Fox et. al., 2013; Johnson et. al., 2013; Warriss et al., 2006), pigs are particularly susceptible to the effects of environmentally-induced heat stress (HS), which can result in production losses as well as morbidity and mortality (Johnson et al., 2015). Although the recommended temperature range for finishing and market weight pigs is 10 to 25°C (FASS, 2010), pigs are often exposed to temperatures outside this range. In addition to HS, other stressors such as handling may increase heat production (Stewart et al., 2005) resulting in a greater body temperature and a subsequent increase in HS susceptibility (Addis et al., 1967). Therefore, elevated body temperature could be a potential tool to determine whether swine are stressed in a commercial setting (Brown-Brandl et. al., 2003; Kammergaard et al., 2013).

The study objective was to determine if infrared thermography could be used as a non-invasive, hands-off approach to accurately monitor the welfare of swine by comparing changes in skin surface temperature (SST), core body temperature (CBT), behavior, and heart rate during acute heat stress (AHS) and subsequent recovery in thermoneutral (TN) conditions. We hypothesized that skin temperature in peripheral regions (ear, ear base) would be better suited to predict CBT than trunk regions (neck, shoulder, thorax, flank, rump, ham).

Materials and methods

2.1. Animals and study design

All experimental procedures using animals were approved by the Purdue University Animal Care and Use Committee (protocol #1406001096). In 8 repetitions (2 pigs per repetition),

16 crossbred pigs (n = 8 gilts (nulliparous female) and 8 barrows (castrated male); 98.2 ± 2.0 kg BW; $\frac{1}{4}$ Large White $\times$ $\frac{1}{4}$ Landrace $\times$ $\frac{1}{2}$ Duroc; 100 d of age) were housed in the Purdue University Swine Farm Environmental rooms and maintained in TN conditions (20.6 ± 0.1 °C and 78.6 ± 0.1 % RH) to acclimate for 7 d. Pigs had *ad libitum* access to feed and water during the acclimation phase. The feed consisted primarily of corn and soybean meal and met or exceeded nutrient requirements (NRC, 2012). Light cycles were set at a 12 h light: 12 h dark cycle and all pigs were individually housed (1.8 m x 1.2 m pens with metal grate flooring) throughout the entire study.

On d 4, 5, and 6 of the acclimation period, pigs from each repetition were handled and individually moved into an identical environmental room across a corridor (6 m) set at 20.6 ± 0.1 °C and 70.6 ± 0.1 % RH for 30 min (this room would later serve as the AHS room) and then returned to their room (to later serve as the TN room). This training occurred at similar times each day at 1000 h and the same groups of pigs were moved each time during acclimation period and during the experiment. The training was provided to minimize the effects of handling when pigs were moved into the AHS room for the experiment. Approximately 90 min prior to experimental AHS at 0800 h on d 7, temperature transmitters calibrated by the manufacturer (CorTempTM T150002; accuracy: ± 0.1 °C; 2.0 cm x 0.5 cm; HQ Inc.; Palmetto, FL) were orally administered to the two pigs to monitor CBT and pigs were feed and water-restricted during the experiment period for 90 min. Pigs were fitted behind the forelimbs with an electrode-containing strap that was telemetrically connected to a receiver watch (Polar RS800CX, Finland) to collect heart rate data. Prior to data collection, pigs were allowed to rest in their home pen for 30 min to minimize any physiological changes due to handling. Baseline CBT and SST were collected every 1 min during the last 5 min of the 30-min rest period. Concurrently, heart rate was

monitored for 5 min and average baseline beats per minute (bpm) was calculated. If the heart rate was higher than 250 bpm and less than 75 bpm, such data were considered noise and not included in analysis. This cleaning of data excluded 6.4 % of total heart rate data.

Data loggers (HOBO; Onset ®; Bourne, MA) were used to record temperature and humidity in TN ($20.6 \pm 0.1^{\circ}\text{C}$; $78.6 \pm 0.2\%$ RH) and AHS ($39.3 \pm 0.1^{\circ}\text{C}$; $38.2 \pm 0.2\%$ RH) rooms. After the 30-min rest period, two pigs were moved into the AHS room and CBT and SST were recorded every 1 min for the first 5 min, and then every 5 min for the final 25 min. After the 30 min of AHS, pigs were returned to their home pen in TN conditions and CBT and SST were recorded as in the AHS room.

2.2. Thermal imaging

Skin surface temperature was determined by taking a broadside photo from a similar distance (1.5 m) and position perpendicular to the camera optical axis of individual pigs using an infrared thermal camera (FLIR-T62101; -20°C to 1200°C temperature range with $\pm 2\%$ accuracy; NETD: $<0.045^{\circ}\text{C}$ @ 30°C ; 320 x 240 pixels infrared resolution; 120° swivel lens and $25^{\circ} \times 19^{\circ}$ field of view with 1-8x continuous zoom; FLIR Systems Inc., Wilsonville, OR) with emissivity set at 1 and photos were analyzed using FLIR Tools software (version 2.1). Because pigs were housed within closed and clean environmental rooms without windows to the outside and water was not available during the experiment, no reflections or infrared radiation could influence measured surface temperatures and pigs remained dry and clean throughout the study.

Temperatures at nine anatomical locations were recorded in each photo. The locations (Fig. 1) included ear (back of pinna), ear base (transition between pinna and neck), neck (above the vertebrae), shoulder (across the scapula), thorax (posterior to elbow over the fore ribs), flank (past the ribs and in front of the stifle muscle), rump (dorsal to hip), ham (below tail and above

hocks), and body average (included the majority of the side profile). Body parts were selected manually using software function of the camera. An area of approximately 2 cm^2 within the parts as shown in Figure 1 was selected to record the skin temperature for all body parts except ear base (approximately 1 cm^2) and body average (approximately 1300 cm^2). When the body parts were covered or enough areas were not exposed, those body parts were considered missing for analysis purpose.

2.3. Behavior observations

Pig behavior was recorded while pigs were in the AHS and TN room using ceiling mounted cameras (Panasonic WV-CP254H, Matsushita Electric Industrial Co. Ltd., Osaka, Japan) attached to the digital video recorder system. Video files were analyzed in Observer XT 11.5 (Noldus, The Netherlands) by one trained individual using continuous sampling technique. Postures included standing (when a pig was up on its four legs without running or jumping), lying (sternum or side in contact with floor), sitting (fore-legs extended, hind legs flexed and sternum off ground), and hyperactive (standing on its four legs but with running and attempts to jump within the pen). Behaviors included attempted feeding (head in feeder), attempted drinking (snout in contact with nipple drinker), activity (sham chewing, moving, chewing on pen objects), and inactive (absence of any of the previously noted behaviors). Percent of time each pig displayed a posture and a behavior was calculated. The 10 possible behaviors and postures were lying and no activity, lying and activity, sitting and no activity, sitting and drinking, sitting and eating, sitting and activity, standing and no activity, standing and drinking, standing and feeding, standing and activity, and hyperactivity. However, pigs did not display the combination of sitting and eating while they were in AHS or TN rooms.

2.4. Statistical analysis

Core body temperature data and skin surface temperature data were analyzed using the PROC MIXED procedure in SAS 9.4(SAS Institute Inc., Cary, NC). Data within the AHS and TN periods were analyzed separately and were conducted with time (0-30 min) as the statistical model component. The statistical model was: $Y_{ijk} = \mu + T_i + e_{ij}$. Where, Y = parameter of interest, μ = mean, T = time, and e = residual. For repeated analyses of time within period, CBT and SST for each pig were analyzed using repeated measures with an auto-regressive covariance structure with time within the AHS or TN periods as the repeated effect. To analyze differences between periods, CBT and SST data within each period were averaged for individual pigs and the statistical model component was period (AHS, TN). The statistical model for period analyses was: $Y_{ijk} = \mu + P_i + e_{ik}$. Where, Y = parameter of interest, μ = mean, P = period, and e = residual. Repetition was used as a random effect and CBT and SST during the pre-treatment period were used as a covariate for all CBT and SST analyses, respectively. Sex was included as a random variable for all analyses; however, because no differences were detected for any parameter it was removed from the final analysis.

Average percent of time spent in displaying a particular posture and behavior while in AHS and TN rooms were compared using general linear model. For data cleaning, only heart rate data between 75 and 250 bpm were considered for analysis. Average heart rates during the 10-min intervals when the pigs were in AHS and TN rooms were compared to baseline heart rate. Results are presented as mean $\pm$ SE where possible and $P < 0.05$ was considered significantly different and $0.05 \leq P \leq 0.1$ was considered to be a tendency.

3. Results

Pre-treatment CBT and SST for each body location are presented in Table 1. During pre-treatment, SST was greater at the ear base ($35.6 \pm 0.2^\circ\text{C}$) compared to all other SST ($P < 0.01$;

Table 1). At the end of the AHS period, SST at the ear, ear base, neck, shoulder, thorax, flank, rump, and ham were increased overall from pre-treatment by $5.5 \pm 0.4^{\circ}\text{C}$, $5.5 \pm 0.5^{\circ}\text{C}$, $6.9 \pm 0.4^{\circ}\text{C}$, $6.9 \pm 0.4^{\circ}\text{C}$, $7.1 \pm 0.4^{\circ}\text{C}$, 6.7 ± 0.4 , $7.1 \pm 0.4^{\circ}\text{C}$, and $6.5 \pm 0.4^{\circ}\text{C}$, respectively (Table 1). During the TN period, ear, ear base, neck, shoulder, thorax, flank, rump, and ham SST were reduced by $1.5 \pm 0.5^{\circ}\text{C}$, $1.4 \pm 0.6^{\circ}\text{C}$, $2.1 \pm 0.5^{\circ}\text{C}$, $2.6 \pm 0.5^{\circ}\text{C}$, $2.2 \pm 0.5^{\circ}\text{C}$, $1.9 \pm 0.6^{\circ}\text{C}$, $1.8 \pm 0.6^{\circ}\text{C}$, and $2.3 \pm 0.5^{\circ}\text{C}$, respectively, compared to baseline (Fig. 2). Following the AHS period, the SST of the different parts of the body did not differ from each other ($P = 0.30$). The effects of AHS on change in SST and CBT from pre-treatment are presented in Fig. 2 and 3. A time effect was detected in SST ($P < 0.01$) and CBT ($P < 0.01$) during the AHS period (Fig. 2). In the TN period, a time effect ($P < 0.01$) was detected for SST (Fig. 2) where average SST during the first min in TN period was $4.0 \pm 0.2^{\circ}\text{C}$ higher than baseline and did not decline to baseline SST within 30 min (Fig 2). A time difference was observed during the AHS period where CBT was increased ($P < 0.01$; 0.9°C) when comparing 1 min to 30 min; however, no CBT time point difference was observed during the TN period ($P = 0.98$; Fig 2). When pigs were moved from AHS to TN treatment SST dropped abruptly and then decreased slowly and remained constant without reaching to the baseline within 30 min in TN room (Figure 2). Overall average and maximum CBT during the TN period was greater ($P < 0.01$; 0.2 and 0.3°C , respectively) compared to maximum CBT during the AHS period (Fig. 3).

Behavioral data during the AHS period are presented in Table 2. When the pigs were exposed to AHS, they tended ($P = 0.10$) to reduce time spent lying with no activity, increase ($P < 0.01$) time spent standing with no activity and standing with activity, and tended to be more hyperactive ($P = 0.06$). Pigs exposed to AHS tended ($P = 0.08$) to spend less time lying with

activity and sitting and attempting to drink. There were no differences ($P > 0.20$) detected in all other behavior and posture combinations.

During the pre-treatment period, average heart rate was 132 ± 5.5 bpm. Overall, no differences ($P > 0.05$) were detected in average heart rate during the AHS (pooled 142 ± 5.9) and TN periods (pooled 143 ± 5.9 bpm; data not shown).

4. DISCUSSION

Methods to accurately and efficiently monitor the welfare of swine are in high demand. While current measures (e.g., blood cortisol, rectal temperature, heart rate) are successful in estimating stress levels, increased stress due to handling and labor costs associated with these approaches may be a deterrent to implementation in commercial pig production. Therefore, it is imperative that new technologies are evaluated for the non-invasive monitoring of swine health and welfare.

Infrared thermography serves as a non-invasive method of measuring skin temperature because temperature can be assessed without restraining the pigs. It offers many advantages for producers and researchers alike, including reduced handling stress, disease spread, and labor. However, factors like ambient temperature (Loughmiller et. al., 2001), anatomical location, and equipment capabilities may limit the accuracy of this technology (Soerensen, 2015). While other studies have detected correlations between rectal temperatures and ear, vulva, and eye surface temperature (Schmidt et al., 2013), collecting temperatures at these locations either require animal handling or approaching animals which can result in increased time, labor, and animal stress.

Pig SST varies with body location, heat source exposure, hair presence, exposure duration, previous HS exposure, subcutaneous body fat, and air circulation (Hales, 1973; Soerensen and Pedersen, 2015). Similarly, CBT varies with environmental temperature, pre-exposure and adaptation to HS, disease condition, and physiological state (primarily in females; Scolari et al., 2011). In accordance with previous reports (as reviewed by Soerensen and Pedersen, 2015), pre-treatment ear base SST was greater compared to other body locations likely due to a greater blood supply and radiative heat exchange. However, SST on ear base was not different from SST on other body parts during AHS. This could be due to increased vasodilation when pigs were exposed to higher temperatures (35 and 37.5 °C) as explained by Stombaugh and Roller (1977). The increase in blood supply and subsequent ear SST during TN treatment may explain the greater correlation with CBT described by Schmidt and colleagues (2013). Acute HS exposure increased SST at all locations and CBT, and subsequent TN exposure reduced SST abruptly at all locations for another 10 min and remained constant without reaching pre-treatment levels for the remaining 20 min of the TN period. This pattern was similar in all nine-body locations measured and in the average of these locations (majority of side profile; Fig 1). This indicated that CBT of pigs continued to increase even after TN exposure followed by AHS exposure.

Despite a reduced SST, pig CBT was increased during the TN period (Fig 3). Greater CBT could lead to intestinal epithelium damage and increase intestinal permeability allowing for LPS leakage resulting in the production of cytokines such as TNF- α and IL-6 (Lambert et al., 2002; Pearce et al., 2012). In addition, this LPS leakage and potential increase in cytokines could have led to illness in the pigs and induced more lying behavior when pigs were returned to the TN environment (Dantzer et al., 2008).

Behaviors could also be used as an indication of welfare and predictor of physiological changes due to stressors. When acutely heat stressed, pigs spent less time lying down compared to the TN period. During AHS, pigs exhibited more restless behaviors and more activity compared to the TN period. This increased activity was likely due to sudden AHS exposure and corresponding restlessness (Peeters et al., 2005); although, these data are in contrast to previous reports (Hicks et al., 1998; Quiniou et al., 2000) indicating that inactivity and lying behavior is associated with increased ambient temperature. However, the aforementioned studies looked at longer HS duration compared to the current study and when pigs were exposed to TN conditions they likely utilized the comparatively cooler metal mesh floor to lie down and lose excessive body heat through conduction.

While it appears that AHS has no effect on heart rate, the present experiment was only conducted over a 30-min period (shorter than most practical applications like transportation). The greatest heart rate increase (0-10 and 31-40 min) were time periods when the pigs were required to walk (approximately 6 m) to a new environment (from TN to AHS room or vice versa); therefore, a greater stress response resulting from handling could explain the observed heart rate increase. Previous studies (Goumon et al., 2013; Lewis et al., 2008) have reported that transportation and exposure to novel experience can increase heart rate. In the current study, heart rate returned to baseline after only a 30 min exposure to TN. A study where pigs were exposed to longer HS duration (Marple et al., 1974) reported that heart rate increased significantly starting 160 min prior to death when pigs were exposed to HS by linearly increasing from 27°C at the rate of 5°C every hour. If the pigs in current study were exposed to HS for a longer duration, an increase in heart rate would likely have been more obvious.

5. Conclusion

In TN conditions, ear base SST could be used to estimate CBT without handling pigs. However, when pigs were moved from AHS to TN conditions, SST dropped drastically within 10 min while CBT remained increased for at least 30 min. Exposure to AHS could result in increased activity and restlessness in pigs, but heart rate was not affected by AHS or by subsequent exposure to TN conditions.

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Fig 1. Location of each infrared measurement are marked as follows: 1. Ear, 2. Ear base, 3. Neck, 4. Shoulder, 5. Thorax, 6. Flank, 7. Rump, 8. Ham, and 9. Body average.

Fig 2. Change in skin surface temperatures from baseline (average of SST taken every minute for 5 min prior to AHS treatment) at each anatomical location during AHS ($39.3 \pm 0.1^{\circ}\text{C}$) and the subsequent TN exposure ($20.6 \pm 0.1^{\circ}\text{C}$). Each graph represents an anatomical location. For all measures, SST increased ($P < 0.01$) during AHS and decreased from this temperature after they were placed in TN ($P < 0.01$). In contrast CBT rose during AHS ($P < 0.01$) and continued to rise during TN ($P < 0.01$).

Fig 3. Core body temperature at pre-treatment ($20.6 \pm 0.1^{\circ}\text{C}$), during 30 min of acute heat stress ($39.3 \pm 0.1^{\circ}\text{C}$; AHS) and 30 min of thermoneutral ($20.6 \pm 0.1^{\circ}\text{C}$; TN) environmental conditions. An asterisk (*) indicates significant differences ($P < 0.01$) between periods. PT=Time period before the actual study started

Table 1: Thermal indices of pigs prior to and following AHS¹

Body parts	T_B²	T_{30 MIN}³	T_{31 MIN}⁴	T_{60 MIN}⁵
Average	33.5 ± 0.2	40.5 ± 0.4	37.7 ± 0.4	35.8 ± 0.3
Ear	34.8 ± 0.3	40.8 ± 0.4	38.4 ± 0.4	36.6 ± 0.4
Ear base	35.6 ± 0.2*	41.3 ± 0.5	38.4 ± 0.4	36.6 ± 0.4
Neck	34.2 ± 0.2	41.0 ± 0.3	38.0 ± 0.4	35.8 ± 0.3
Shoulder	33.7 ± 0.2	40.6 ± 0.4	38.0 ± 0.3	36.1 ± 0.3
Thorax	33.0 ± 0.3	39.9 ± 0.4	37.4 ± 0.5	35.6 ± 0.5
Flank	33.7 ± 0.2	40.1 ± 0.4	37.4 ± 0.4	36.0 ± 0.4
Rump	33.4 ± 0.2	40.4 ± 0.4	37.4 ± 0.5	35.1 ± 0.3
Ham	34.0 ± 0.2	39.9 ± 0.5	37.5 ± 0.5	35.9 ± 0.4
CBT⁶	39.3 ± 0.1	40.3 ± 0.2	40.4 ± 0.2	40.5 ± 0.2
P⁷	<0.01	0.30	0.60	0.07

¹ Acute heat stress² Baseline temperature, °C³ Average temperature of pigs at end of AHS, °C⁴ Average temperature of pigs at first recording after return to thermoneutral room, °C⁵ Average temperature of pigs at end of observations, °C⁶ Core body temperature⁷ P-values were compared only for the SST on the body parts

*SST at baseline on ear base was the warmest (P < 0.01)

Table 2. Average percent of time pigs displayed combinations of behavior and posture in AHS¹ and TN² rooms

Behavior	AHS¹	TN²	SEM³	P
Lying and no activity	35.3	50.1	6.2	0.10
Lying and activity	9.3	17.6	3.1	0.08
Sitting and no activity	3.1	0.9	1.3	0.20
Sitting and attempted drinking	0.2	2.1	0.7	0.08
Sitting and activity	1.2	4.3	2.1	0.30
Standing and no activity	3.4	0.1	0.8	0.007
Standing and attempted drinking	2.1	1.6	0.4	0.50
Standing and attempted feeding	1.7	2.6	1.0	0.40
Standing and activity	40.2	20.7	4.6	0.007
Hyperactivity	3.5	0.0	1.2	0.06

¹ 30-min period of acute heat stress (39.4 °C)

² 30-min period in thermoneutral room (20 °C) after heat stress

³ Standard error of the mean