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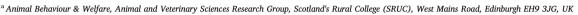
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Infrared thermography of agonistic behaviour in pigs

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ABSTRACT

Infrared thermography (IRT) or thermal imaging is increasingly being used as a non-invasive method to gain information on animals' physiological and emotional state. IRT has the potential to serve as a non-invasive quantitative assessment method but few studies have examined its utility in predicting welfare-relevant outcomes of dynamic scenarios relevant to commercial farming. This study used 1284 thermal images taken from 46 pigs in a controlled test environment while they engaged in an agonistic encounter (dyadic contest) at 13 wk. of age. Images were taken of the complete body from a dorsal perspective. A pilot study indicated that a rectangular thermal window on the back region was the most suitable and reliable area for obtaining temperature data in this situation. From this thermal window, the average, minimum and maximum temperature, standard deviation and coefficient of variation (CV) were obtained. These were analysed in relation to contest phase (from noncontact assessment, through escalated fighting to retreat), fight occurrence, contest duration, contest outcome (winner/loser status) and changes in blood glucose, blood lactate, and skin injuries. Variables showed a strong change in response to the moment of contest resolution (retreat of the loser); temperatures reduced sharply and CV increased, but did not differ between winners and losers. Contests that included a fight showed lower temperatures. Contest duration, body weight and sex only had minor influences on the temperatures. As the drop in temperature at contest resolution was irrespective of contest intensity, and the pattern was similar in winners and losers, this data potentially reflects vasoconstriction as a result of psychological stress rather than solely a physiological change. The study shows that peripheral temperature, as recorded by IRT, responds to the intensity and phases of a contest and may allow new insight into the physiological and welfare outcomes of aggressive behaviour.

1. Introduction

Infrared thermography (IRT) or thermal imaging is a non-invasive method that uses specialised imaging cameras to capture infrared (heat) radiation emitted from the surface of an object. It then produces a thermograph that allows visualisation and quantification of the temperature distribution [13,35]. This thermograph or thermogram can be used to detect physiological and behavioural changes in endotherm animals [18].

In the animal and welfare science fields, IRT has initially been applied as a method to measure changes in body temperature in response to injury, disease, handling, transportation, and fatigue [20]. As such, IRT as a health and welfare assessment tool has been used in a wide variety of animals including livestock, companion and wild animals

[7,11,23]. More recent studies have also used IRT to make inferences about the individual's emotional state, in particular in response to situations likely to cause stress, rather than rely on more traditional physiological measures. For example, IRT has been used to assess the relationship between facial surface temperature and emotional states in rhesus monkeys concluding that a decrease in nasal temperature was correlated with a negative emotional state [24]. Similar approaches have thereafter been applied to other non-human species and circumstances (primates: [6,15]; dogs: [33]; rabbits: [17]). The use of IRT to study emotions has been more widely used in humans [8], for example to study emotions in infants (e.g. [10]).

IRT has been applied to treatment comparisons in which animals have been exposed to an acute stress treatment, but its sensitivity in detecting individual differences in behavioural and physiological

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response to stress has only been sparsely tested. The objective of this study was to test whether peripheral temperature as measured by IRT is associated with variations in behavioural and physiological responses of pigs in a situation with strong emotional valence and arousal, namely agonistic encounters. More specifically, we hypothesized that IRT response is associated with the physiological and behavioural changes occurring during dyadic contests. In addition, given the potentially different emotional state in winners and losers [3], we also hypothesize that contest outcome will influence IRT responses. Pigs were selected as an ideal species since hair coverage is minimal and IRT studies have identified different thermal windows that correlate to some extent with core body temperatures [28]. The pig body, rather than only the eye, is therefore very suitable for IRT. Furthermore, aggression between pigs is a common occurrence on farms and is a considerable animal welfare issue [25]. Obtaining insight into this welfare issue through a non-invasive measure may improve understanding of the physical and potentially emotional outcomes of aggression and of winning and losing.

2. Material & methods

This research was part of a larger trial on aggression in pigs and all tests were carried out for the purpose of multiple research questions. The project was carried out in accordance with the European Guidelines for accommodation and care of animals, the ASAB/ABS and ARRIVE guidelines and UK Government DEFRA animal welfare codes. The project received ethical approval by SRUC's Animal Ethics Committee (ED AE 40-2014) and the UK Government Home Office under the Animals Scientific Procedures Act 1986.

2.1. Animals and housing

In total, 46 pigs with suitable thermal image data were sampled out of a population of 134 growing pigs (Large White \times Landrace dam \times American Hampshire sire). Entire males (not castrated) and females were studied over two batches at the SRUC pig research unit (Easter Howgate, UK). Tail and teeth were kept intact. Pigs were housed by litter group in solid floor, straw bedded pens measuring $1.9\times5.8~\text{m}$ (ca. $1.0\text{-}1.1~\text{m}^2/\text{pig}$) which were cleaned and replenished daily. Pigs had ad libitum access to water and dry pelleted feed suitable to their age and growth. The room temperature was kept at $16.1~\pm~0.3~\text{C}$ with a humidity of $53.5~\pm~0.1\%$ using Dicam© monitoring and ceiling vents and heat sources. From 5 to 7 weeks of age the pigs were gradually habituated human handling and the test situation, including ear touching, weighing, time spent in the sampling crate, movement to the contest staging area (but not the contest arena itself) and diminishing group size in order to reduce potential fearfulness.

2.2. Test situation

Thermal images (explained in section 'Thermal images') were captured during 69 dyadic contests between pigs. Contests were staged between two pigs who were unknown to each other, were all 13 weeks of age, and had a weight difference varying between 0 and 20%. Dyads were composed of male-male, female-female, and male-female contests. Contests were held in a solid sided arena measuring $2.9 \times 3.8 \, \text{m}$ with a solid floor covered by a thin layer of wood shavings (Fig. 1). At either side of the arena were $2.9 \, \text{m} \times 1.9 \, \text{m}$ holding pens to temporarily house each pig prior to the test starting. The room was lit by nine overhead fluorescent strips providing 80-110 lx, and no natural light could enter. The room in which the arena was situated had an ambient temperature of on average 16.1 \pm 0.03 °C (range 15–17.5 °C) with a humidity of $53 \pm 0.1\%$ (range 50–58). The contest started as soon as the two pigs entered the arena simultaneously from opposite sides. A contest was terminated when a) a winner was apparent (loser retreats and does not retaliate within 1 min after retreat); b) after 20 min (time-out); or c) due to an end-point such as fear behaviour or mounting. The total contest



Fig. 1. Opponents showing agonistic behaviour in the contest arena. The silver dot on the rear of the left pig can be distinguished on a thermal image due to difference in emissivity, and therefore enables individual identification. Copyright: M. Farish.

duration and the fight duration (time spent in mutual aggression) were recorded for this study.

2.3. Blood glucose, blood lactate and skin lesions

Blood values were measured within 2 min before and within 5 min after the contest by obtaining a drop of blood from the ear vein of each pig. This was done using a small flat blade lancet to make a tiny prick in the ear vein while the pig was standing in a sampling crate. The drop of blood was directly applied to a blood glucose meter (iDia monitor; IME-DC, Germany) and a blood lactate meter (The Edge; Apex Biotechnology Copr, Taiwan). The meters instantly provide a read out value in mmol/ml. The measurement range for the glucose meter was 0.6–33.3 mmol/ml and for the lactate meter 0.7–22.3 mmol/ml. Monitors were calibrated before each session with the appropriate calibration fluids. The pre-contest value was subtracted from the post-contest value to obtain the change in glucose and lactate which was used for further analyses.

Skin lesions, scratches on the skin due to the receipt of bites during an aggressive encounter, were counted by an experienced single observer in the home pen pre-contest and directly after the contest in the holding pens. The number of fresh skin lesions (bright red, with no scab formation) was counted separately for the front, middle and rear of the body on the left and right side. The number of skin lesions pre-contest was subtracted from the number post-contest to obtain the actual number of lesions gained during the contest.

2.4. Thermal images

A FLIR SC620 thermal camera with wide angle lens $(45^\circ \times 34^\circ/0.1\,\text{m})$ was used to collect thermographs at $10\,\text{s}$ intervals for the full duration of the contests. To distinguish the two pigs on the thermographs, a silver mark (Aluspray®, an aluminium wound spray for veterinary purposes) was sprayed on the back at the base of the tail of one of the two pigs before entry into the arena (Fig. 1). Due to the different emissivity of the spray compared to the skin (pig skin emissivity 0.94-0.98; [27]) the spray mark showed up differently on the thermograph despite having the same temperature. The rear of the pig was chosen as this area receives fewest bites and would therefore have least chance of fading during the course of a fight. The thermal imaging camera was positioned, approximately 5 m above the ground at an angle of 30° , using a Manfrotto® adjustable arm and clamp, providing an overhead view of the full contest arena. A Kestrel© 4000 weather meter was used to log the atmospheric temperature and relative

Table 1
Location of the highest temperature observed over 533 thermal images of 36 pigs.

Location	N observations	%	
Ear	338	63.4	
Back	113	21.2	
Rear	44	8.3	
Spine	15	2.8	
Hind leg	20	3.8	
Front leg	3	0.6	

humidity (RH) of the contest room with changes $> 1\,^\circ\text{C}$ or 1% RH inputted to the object parameters throughout the sampling period. The distance between the camera and the pigs was constant throughout, emissivity set to 0.98, and reflective temperature 15–20 $^\circ\text{C}$.

2.5. Selection of the region of interest (ROI)

A pilot study was performed to determine the best area of interest, using a sub sample of 533 images from 36 pigs. Using FLIR ThermaCAM Researcher 2.10 Pro (FLIR Systems, Inc.), a polygon was drawn around the whole pig. From the polygon, the location of the pixel with the highest temperature was assessed. To do this the thermal scale was modified by focusing on the highest temperatures until only one visible spot remained. Then, a mark was placed on this pixel using the function 'flying spot meter'. The location of the spot was then identified by readjusting the image to the normal thermal scale. The location could be categorized into one of the body regions: ears; eyes; back (excluding spine); spine; rear; front leg; and hind leg. Observations on the eye (n = 3) were excluded because of poor visibility from the aerial view. The results, provided in Table 1, show that the ear most often had the highest temperature, followed by the back. Due to the difficulty of imaging the ear during active motion of the pigs (e.g. during a fight), the possibility of saliva and blood on the ear due to bites, and in this case extra blood flow due to blood sampling from the ear, it was decided that the ear was not feasible nor reliable under the current conditions. Therefore, the back region was chosen as the ROI. Previous research has also suggested that this ROI could be a thermal window in piglets [30]. Thermal windows are characterised as heat exchange body areas, perfused with blood and operating as windows into core body temperature [20,22,28].

2.6. Extraction of data from thermal images

Contests with < 17 raw images (n = 29 contests) commonly had too few usable images and were therefore excluded. Contests with > 80 images (~ 3 times the duration of an average contest; n=14 contests) were also excluded as the behavioural repertoire in these contests typically deviated too much compared to the average contest. One contest was excluded due to low image quality (low in pixels). Images were excluded when the two pigs appeared on the image as merged (e.g. because of mounting) or when the image was not sharp due to fast movements. Eventually, from the 67 contests 23 were analysed, resulting in 1284 suitable images for 46 pigs. Of the 46 pigs, 37% were female (n=17) and 63% were male (n=29). Dyads were composed of 48% males against males (n=22), 33% females against males (n=16), and 19% females against females (n=8). Of the 1284 images 50.2% belonged to winners (n=645) and 49.8% to losers (n=639).

In the software FLIR ThermaCAM Researcher 2.10 Pro (FLIR Systems, Inc.) images were set to the 'rain900' pallet for colouration (from warm to cold: white, red, orange, green, blue, dark blue). The polygon tool was used to isolate the ROI. Specifically, a rectangular area (Fig. 2) was selected on the back of each pig, starting just behind the shoulder blades and ending at the start of the rump. The area did not include the belly as this appears as a warmer area due to the organs



Fig. 2. Screenshot of a thermal image from the software *FLIR ThermaCAM Researcher 2.10 Pro.* The rectangular area on the back of the pig marks the region of interest (ROI). On the rear of the right pig, at the base of the tail, the silver spray mark for identification can be seen. Copyright: SRUC.

being located closer to the skin. The area excluded any object from the environment and excluded cool edges (i.e. lines around an ellipse shaped figure that give a lower radiation value due to an angle and therefore do not reflect temperature; for example in Fig. 2 the cool edge is the yellow-blueish line around each pig). Each ROI had between 4000 and 5000 pixels, depending on the orientation of the pigs. The minimum, average, and maximum surface temperature and the standard deviation for each ROI were extracted. For each image the time (hh:mm:ss) was noted to assess the temperature change over time. Henceforth, "temperature" refers to skin surface temperature unless otherwise stated as environmental.

Temperature was assessed for three key time points and two phases, providing five temperature values per animal: 1) start of the contest (first image in which the pig was fully visible); 2) agonistic phase (average during the phase in which agonistic behaviour was shown); 3) retreat (single image closest to the moment that the loser signalled its final submission to the winner; 4) post-retreat phase (average of the images after retreat until the last image), and 5) end of the contest (last image, 1 min after retreat).

2.7. Data analysis

Data were analysed with SAS 9.3 (SAS Institute Inc., Cary, NC, USA). Each pig was considered as statistical unit (n=46), but was nested within dyad to correct for dependence between two pigs within the same contest. Pearson correlations were used to assess relationships between the various contest measures (skin lesions, contest duration, fight duration, glucose and lactate), and between these measures and temperature variables averaged by pig. The contest measures correlated moderately with each other (r=0.33–0.63, average r=0.51). To reduce redundancy in the subsequent modelling, only skin lesions and contest duration were retained as they showed the strongest correlations with the other variables.

The dependent variables were the average temperature (avgT), minimum temperature (minT), and maximum temperature (maxT), recorded for each image. To measure the degree of variation in temperature, the Coefficient of Variation (CV) was analysed as a dependent variable. These dependent variables were used in a generalized linear

model (MIXED Procedure). Initially, all explanatory variables were included in the model. These were: contest outcome (winner/loser), sex (male/female), gender composition of the dyad (MM/FF/MF), body weight at 13 weeks age, weight difference between the opponents, contest duration, number of skin lesions, fight (yes/no), timing (first image/average of the agonistic phase/retreat/average of the post-retreat phase/last image), and test date (5 dates). Variables with the highest non-significant P-value were step-wise removed from the model and the model was reassessed for goodness of fit using the Akaike information criterion (AIC) and Bayesian information criterion (BIC). The variable was omitted only if this improved model fit (lower AIC and BIC). The random effects included pig ID nested within dyad (to indicate that the two opponents in a contest were not independent), and batch number (batch 1 or 2) and pig ID was specified as subject to account for repeated observations per animal (resulting in 46 subjects with five observations each, one for each time point in the contest). The four models showed a normal distribution of the residuals. Values are presented as LSmeans with SE. A P-value < .05 was considered significant, whereas *P*-values > .05 but < 0.10 were reported with their exact value as tendencies.

3. Results

The average temperature (avgT) was $31.6\,^{\circ}\text{C} \pm 0.3$ (range 28.0--34.2) with a coefficient of variation (CV of $2.75\% \pm 0.03$ (0.91–7.39%). The mean minimum temperature (minT) was $28.0\,^{\circ}\text{C} \pm 0.5$ (16.4–32.2) and the maximum temperature (maxT) $33.5\,^{\circ}\text{C} \pm 0.2$ (30.7–35.9). The date of observation did not significantly influence these variables.

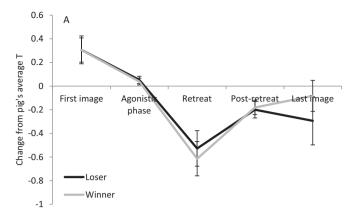
3.1. Temperature changes over time

The avgT, maxT and minT all showed a sharp and significant reduction at the moment of contest resolution, when the loser began to retreat from the winner by making a head-tilt movement (Table 2). After this retreat, the average temperature slightly increased again until the contest was ended one minute later. The contrast is stronger when the moment of retreat is reflected as the change from an individual's average temperature (Fig. 3).

3.2. Contest intensity

The contest duration was on average 7 min (424 \pm 0.5 s), with an average fight duration of 55 \pm 1.7 s. Contest duration and fight duration were unrelated to the avgT, maxT, minT, and CV.

Not all contests involved a fight as some were resolved merely by a unilateral attack from the winner (73% of the contests included a fight, 27% did not). The duration of contests with a fight was on average longer than contests without fight (with fight: 450 \pm 6 s; without fight: 354 \pm 9 s; P<.001). Temperatures did not significantly differ between contests with or without a fight, which may be partly due to the large SE in the contests without a fight as the number of contests in this category were few (Fig. 3b). Fig. 3 reveals that the drop in temperature during retreat is mainly related to contests with a fight, as the different



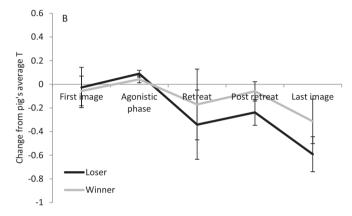


Fig. 3. Average change from each individual's average temperature at various contest stages for contests A) with a fight (n = 32 pigs: 16 winners and 16 losers) and B) without a fight (n = 14 pigs: 7 winners and 7 losers).

contest phases only tend to differ from each other in contests without a fight (winners and losers analysed together: $F_{4,49} = 2.45$; P = .058). In contests without a fight a significant reduction in temperature was only observed in the last image as compared to the first image (P = .03) and the agonistic phase (P = .006), and the value of the last image tended to be lower than in the post-retreat phase (P = .09) (Fig. 3b). The temperature at retreat tended to be lower than the temperature in the preceding agonistic phase (P = .07).

3.3. Physiological parameters and skin lesions

Pigs accumulated on average 72 \pm 10 skin lesions due to the receipt of bites. Temperatures were significantly related to the number of skin lesions on the pig's body. Animals with many skin lesions had a lower minT ($F_{1,172}=4.68; P=.03$) and a greater CV ($F_{1,171}=21.92; P<.001$), which was also clear from the initial correlations (Table 3). The number of skin lesions did not significantly relate to the avgT ($F_{1,172}=2.55; P=.11$) and maxT ($F_{1,172}=0.01; P=.93$).

Table 2
Temperatures (in $^{\circ}$ C) and coefficient of variation (CV; in $^{\circ}$) at different contest stages (n=1284 images). The first image in the contest, the image at retreat of the loser, and the last image are values from a single image whereas pre and post reflect the average of all images in between the fixed time points, i.e. the agonistic phase and the post-retreat phase. Values are LSmeans.

Temperature	First	Pre	Retreat	Post	Last	F statistic	P-value
Minimum Average Maximum CV (%)	29.12 ^a 31.85 ^a 33.43 ^a 2.31 ^a	28.89^{a} 31.73^{a} 33.42^{a} 2.48^{a}	27.98 ^b 31.20 ^b 33.13 ^b 2.95 ^b	28.19^{b} 31.43^{c} 33.46^{a} 3.03^{b}	29.10 ^b 31.39 ^{bc} 33.39 ^a 2.97 ^b	$F_{4,172} = 14.41$ $F_{4,172} = 14.28$ $F_{4,171} = 3.86$ $F_{4,171} = 17.11$	< 0.001 < 0.001 0.005 < 0.001

Table 3 Correlations between fixed effects and temperature variables averaged by pig (n = 46).

	Lesions	Contest duration	Fight duration	$\Delta Glucose$	ΔLactate
AvgT MaxT MinT CV	-0.44* -0.27 -0.51*	- 0.25 - 0.02 - 0.40* 0.50*	-0.03 0.10 -0.23 0.38	-0.35 -0.36 -0.32	-0.22 -0.19 -0.49*

^{*} P-value < .001.

Blood glucose and lactate increased on average during the course of the contest (glucose: 1.04 ± 0.22 mMol/L increase (min-max: 3.2–10.9); lactate: 5.8 ± 0.87 mMol/L increase (min-max: 0.6–22.3). The change in blood glucose and blood lactate did not differ between winners and losers (both P > .10) and was unrelated to the temperature values, except that a relative increase in blood lactate related to a lower minT ($b = -0.073 \pm 0.036$ °C per mMol/L increase; $F_{1.171} = 4.09$; P = .04).

3.4. Individual characteristics

Winners and losers did not significantly differ in avgT, minT, maxT or CV (all P > .10). They also did not differ in their temperatures depending on the stage of the contest. For example, winners and losers had a similar temperature at the moment of retreat (Fig. 3).

The CV tended to be lower in pigs with a higher body weight $(b=-0.028\%/\mathrm{kg};\ F_{1,171}=2.91;\ P=.09)$ but body weight did not relate to avgT, minT or maxT (all P>.10). A larger weight difference between the two opponents increased the minT $(b=0.04\,^{\circ}\mathrm{C/kg};\ F_{1,171}=5.75;\ P=.02)$ and tended to reduce the CV $(b=-0.016\%/\mathrm{kg};\ F_{1,171}=3.53;\ P=.06)$ as compared to pigs in dyads of similar body weight. Body weight difference did not influence the avgT $(F_{1,172}=2.50;\ P=.12)$ and maxT $(F_{1,172}=1.75;\ P=.19)$. Sex and the gender composition within a contest (MM/FF/MF) did not influence any of the observed temperature variables (all P>.10).

4. Discussion

The use of infrared thermography (IRT) for assessing animal welfare shows promise but is still in development. To date, IRT has been shown to associate with acute stressors with an indication that at least part of the thermal response reflects the emotional reaction to the stressor in addition to changes in activity, physiology and metabolism (rabbits: [17]; dogs: [33]; cattle: [29]; horses: [34]). However, the association between peripheral temperature and individual differences in stress response has only been marginally explored. Furthermore, the sensitivity of peripheral temperature to time-dependent phases of a highly dynamic stress situation has not been examined. Here we made a step forward by assessing over one thousand IRT images of pigs during an agonistic encounter and examining how temperature was related to individual behaviour and contest outcomes and how sensitive the temperature changes were to the phase of the contest. The main determinant of the peripheral temperature in an ambient temperaturecontrolled environment was the moment at which the loser retreated. Other parameters such as sex and body weight only played a minor role. Whilst the number of skin lesions was associated with temperature changes, other aspects of the contest situation were not, including the occurrence of an escalated fight, the duration of this fight and the contest as a whole, the change in blood glucose and lactate and whether the animal was a winner or loser. This strengthens the overall concept that the reduction in temperature at the moment of retreat of the loser is only partially related to the physiological intensity of the fight, and is in addition potentially influenced by psychological factors such as emotional state.

Mitchell [20] suggests that infrared thermography applications can

be divided into four broad functional categories: a) for monitoring changes in body temperature due to environmental pressures such as high ambient temperature; b) for monitoring the effects on body and skin surface temperature due to transport and handling; c) for monitoring injuries, pathology and inflammatory responses and; d) for monitoring the cardiovascular responses to a stressful event or state. We discuss the current results in light of the two latter applications.

4.1. Injury and fatigue

Injury involves a local temperature increase and can be detected by IRT. During reciprocal aggression pigs deliver bites mainly to their opponents' front side of the body (head and shoulders) including the ears [31]. The underlying metabolic vasoconstriction or vasodilatation that acts as thermoregulation mechanisms may also influence body surface temperature of other areas. Skin injuries itself, which are easily detected from distance on a normal RGB camera, are in general not detectable on the infrared image unless skin injuries are numerous and bleeding. However, the temperatures on the ROI, the dorsal area, did relate to the number of skin lesions. In fact, skin lesions were a better predictor of skin temperature than contest duration or fight duration. Pigs with many lesions showed a reduction in skin temperature rather than an increase. Before the onset of an inflammatory response, which would result in an increase in temperature, there may first be a cooling of the surface temperature due to evaporation from fluids on the skin such as blood from the wound and saliva from the biter (e.g. [18]). Also a temporary capillary constriction may occur at the injury site. Moreover, skin lesions correlate with contest duration, and thus more skin lesions indicate a longer time spent in the contest arena. In this particular situation, the temperature in the contest arena was lower than the temperature in the home pen causing pigs to gradually cool down. As most contests were short the skin lesions did show a stronger relationship to the IRT than the contest duration.

IRT can be used to detect muscle fatigue [1], which is also reflected in lactate. An increase in blood lactate here related to a lower minimum temperature. Fatigue, reflected by the increase in lactate, may have resulted in lower surface temperatures due to associated physiological and psychological stress (e.g. [17,24]). Other than the abovementioned, the thermal images did not reveal differences in the animal's change in blood glucose or blood lactate level.

4.2. Psychological response

The most prominent finding in this study is the sharp drop in temperature at the moment of contest resolution, i.e. the loser's retreat. Existing studies have demonstrated a link between reduced surface temperature and a negative emotional state (e.g. [17,24]). Moe et al. [21] also found a drop in temperature during a positive emotional state, relating the decreased temperature to emotional arousal rather than emotional valence.

When unfamiliar pigs encounter they need to establish a dominance relationship [19]. The subordinate pig signals its submission by a sharp turn away from the opponent, also called a head-tilt [14]. The time of the final head tilt was recorded in each contest and the thermal image within 5 s of the head-tilt was used to reflect this moment. In contests with a fight the temperature is reduced compared to the agonistic phase beforehand, and a similar reduction was found to be a tendency in the contests without a fight. This shows that even in the contests without much physical activity (i.e. no fight), and therefore also without much skin lesions, there was a thermal response to the moment that the contest outcome was settled, with both winners and losers showing a drop in temperature. However, we interpret this finding with some caution given that the results of contests without a fight are based on a limited sample size. Social defeat is well-known to be a powerful stressor [2]. In other IRT studies of acute stress, including social stress, a decrease in surface temperature was also observed [17,29].

Furthermore, repeated social defeat has been shown to induce chronic hyperthermia in rats' core body temperature [16]. In this condition, it could cause long term temperature modification in response to a mild stress. Commercial pigs are regularly regrouped during the production phases [25] and therefore the possibility of negative effects of repeated social defeat on their thermoregulation should not be disregarded. However, winners and losers did not significantly differ in their thermal response at any phase during the contests, including the moment of retreat. This may imply that the moment of defeat equally triggers a response in the winner as in the loser. Although winners and losers show different emotional responses after the fight as assessed through Qualitative Behaviour Assessment (pigs: [3]), the moment of retreat might indicate a climax in physical and psychological effort in which both opponents compete to their maximum capacity.

Whereas in contests with a fight the temperature increased again after retreat, the temperature in contests without fight significantly decreased towards the end of the contest. This might be due to a higher level of bullying behaviour (16% more) in contests that are concluded without a fight [4]. Bullying, in which the loser is being chased by the winner, is by farmers perceived to be more stressful for the pigs than the actual fight [26]. Less aggression at the first encounter may result in more aggression at a later stage [32] and this may thus result in more chronic stress.

These observations suggest that the physical effort committed to the contest was not the primary driver of peripheral temperature changes, suggesting that injury and the emotional response to stress might have been responsible for the drop in temperature during the contest.

4.3. Selection of the region of interest

For the measurement of emotions the eyes are often the targeted body area (e.g. [29]). Imaging pig eyes does have its difficulties due to the small surface and eye lashes obstructing the image. Moreover, during situations of aggression and pain pigs may naturally squint their eyes [5,9]. Here we have demonstrated that in pigs the dorsal plane can also be imaged for assessing responses to strong stressors such as agonistic encounters. This has huge benefits as it is more feasible to image the dorsal plane from overhead cameras. A similar conclusion was made earlier by Tabuaciri et al. [30] when imaging piglets. For future applications, data from thermal images could be extracted with algorithms that detect the shape of the animal [12], as has been done for automatic recording of behaviour. Combing novel technology, for example using tracking to obtain data and algorithms to extract data, will facilitate the use of IRT to assess physiological and emotional responses to various welfare conditions in livestock.

5. Conclusions

Using infrared thermography, a drop in temperature was observed during agonistic encounters between pigs at the moment of retreat of the loser. The drop in temperature, as compared to the agonistic phase, also tended to be present in contests without a fight, indicating that the thermal response was not solely due to physical activity or skin injuries. The data suggest that the moment of retreat, or contest resolution, is the most stressful moment of the encounter and indicates a similar arousal in both the winner and loser. The results show that in pigs the dorsal plane can be used to detect responses in surface temperature to stressful situations. Imaging the dorsal plane rather than the eye or ear greatly facilitates the ease of using IRT for research and practice, and can be used as a non-invasive measure to obtain additional measures of welfare.

Declaration of Competing Interest

None.

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References

- P. Bartuzi, D. Roman-Liu, T. Wiśniewski, The influence of fatigue on muscle temperature, Int. J. Occup. Saf. Ergon. 18 (2) (2012) 233–243.
- [2] R.J. Blanchard, C.R. McKittrick, D.C. Blanchard, Animal models of social stress: effects on behavior and brain neurochemical systems, Physiol. Behav. 73 (3) (2001) 261–271.
- [3] I. Camerlink, M. Peijnenburg, F. Wemelsfelder, S.P. Turner, Emotions after victory or defeat assessed through qualitative behavioural assessment, skin lesions and blood parameters in pigs, Appl. Anim. Behav. Sci. 183 (2016) 28–34.
- [4] I. Camerlink, G. Arnott, M. Farish, S.P. Turner, Complex contests and the influence of aggressiveness in pigs, Anim. Behav. 121 (2016) 71–78.
- [5] I. Camerlink, E. Coulange, M. Farish, E.M. Baxter, S.P. Turner, Facial expression as a potential measure of both intent and emotion, Sci. Rep. 8 (1) (2018) 17602.
- [6] H. Chotard, S. Ioannou, M. Davila-Ross, Infrared thermal imaging: positive and negative emotions modify the skin temperatures of monkey and ape faces, Am. J. Primatol. 80 (5) (2018) e22863.
- [7] J. Cilulko, P. Janiszewski, M. Bogdaszewski, E. Szczygielska, Infrared thermal imaging in studies of wild animals, Eur. J. Wildl. Res. 59 (1) (2013) 17–23.
- [8] J. Clay-Warner, D.T. Robinson, Infrared thermography as a measure of emotion response, Emot. Rev. 7 (2) (2015) 157–162.
- [9] P. Di Giminiani, V.L. Brierley, A. Scollo, F. Gottardo, E.M. Malcolm, S.A. Edwards, M.C. Leach, The assessment of facial expressions in piglets undergoing tail docking and castration: toward the development of the piglet grimace scale, Front. Vet. Sci. 3 (2016) 100.
- [10] G. Esposito, J. Nakazawa, S. Ogawa, R. Stival, D.L. Putnick, M.H. Bornstein, Using infrared thermography to assess emotional responses to infants, Early Child Dev. Care 185 (3) (2015) 438–447.
- [11] S. Foster, C. Ijichi, The association between infrared thermal imagery of core eye temperature, personality, age and housing in cats, Appl. Anim. Behav. Sci. 189 (2017) 79–84.
- [12] N.H. Franco, A. Gerós, L. Oliveira, I.A.S. Olsson, P. Aguiar, ThermoLabAnimal a high-throughput analysis software for non-invasive thermal assessment of laboratory mice, Physiol. Behav. 207 (2019) 113–121.
- [13] R. Gade, T.B. Moeslund, Thermal cameras and applications: a survey, Mach. Vis. Appl. 25 (1) (2014) 245–262.
- [14] P. Jensen, An analysis of agonistic interaction patterns in group-housed dry sows: aggression regulation through an "avoidance order", Appl. Anim. Ethol. 9 (1) (1982) 47–61.
- [15] F. Kano, S. Hirata, T. Deschner, V. Behringer, J. Call, Nasal temperature drop in response to a playback of conspecific fights in chimpanzees: a thermo-imaging study, Physiol. Behav. 155 (2016) 83–94.
- [16] A.J. Keeney, S. Hogg, C.A. Marsden, Alterations in core body temperature, locomotor activity, and corticosterone following acute and repeated social defeat of male NMRI mice, Physiol. Behav. 74 (1–2) (2001) 177–184.
- [17] N. Ludwig, M. Gargano, F. Luzi, C. Carenzi, M. Verga, Applicability of infrared thermography as a non invasive measurements of stress in rabbit, World Rabbit Sci. 15 (4) (2007) 199–206.
- [18] F. Luzi, M. Mitchell, C. Nanni, V. Redaelli, Thermography: Current Status and Advances in Livestock Animals and in Veterinary Medicine, Thermography: Current Status and Advances in Livestock Animals and in Veterinary Medicine, Fondazione Iniziative Zooprofilattiche e zootecniche, Brescia, Italy, 2013 (ISBN 9788897562061).
- [19] G.B. Meese, R. Ewbank, The establishment and nature of the dominance hierarchy in the domesticated pig, Anim. Behav. 21 (2) (1973) 326–334.
- [20] M.A. Mitchell, Thermal imaging: thermoregulation in relation to animal production and welfare, in: F. Luzi, M.A. Mitchell, L. Nanni Costa, V. Redaelli (Eds.), Thermography: Current Status and Advances in Livestock Animals and in Veterinary Medicine, 2013 Fondazione Iniziative Zooprofilattiche e Zootecniche-Brescia, Brescia, Italy, 2013, pp. 147–162.
- [21] R.O. Moe, S.M. Stubsjøen, J. Bohlin, A. Flø, M. Bakken, Peripheral temperature drop in response to anticipation and consumption of a signaled palatable reward in laying hens (Gallus domesticus), Physiol. Behav. 106 (4) (2012) 527–533.
- [22] J.P. Mortola, Thermographic analysis of body surface temperature of mammals, Zool. Sci. 30 (2) (2013) 118–125.
- [23] I.A. Nääs, R.G. Garcia, F.R. Caldara, Infrared thermal image for assessing animal health and welfare, J. Anim. Behav. Biometeorol. 2 (3) (2014) 66–72.
- [24] K. Nakayama, S. Goto, K. Kuraoka, K. Nakamura, Decrease in nasal temperature of

- rhesus monkeys (Macaca mulatta) in negative emotional state, Physiol. Behav. 84 (5) (2005) 783–790.
- [25] R.S. Peden, S.P. Turner, L.A. Boyle, I. Camerlink, The translation of animal welfare research into practice: the case of mixing aggression between pigs, Appl. Anim. Behav. Sci. 204 (2018) 1–9.
- [26] R.S. Peden, I. Camerlink, L.A. Boyle, F. Akaichi, S.P. Turner, Farmer perceptions of pig aggression compared to animal-based measures of fight outcome, Animals 9 (1) (2019) 22.
- [27] D.D. Soerensen, S. Clausen, J.B. Mercer, L.J. Pedersen, Determining the emissivity of pig skin for accurate infrared thermography, Comput. Electron. Agric. 109 (2014) 52–58.
- [28] D.D. Soerensen, L.J. Pedersen, Infrared skin temperature measurements for monitoring health in pigs: a review, Acta Vet. Scand. 57 (1) (2015) 5.
- [29] M. Stewart, A.L. Schaefer, D.B. Haley, J. Colyn, N.J. Cook, K.J. Stafford, J.R. Webster, Infrared thermography as a non-invasive method for detecting fearrelated responses of cattle to handling procedures, Anim. Welf. 17 (4) (2008) 387–393.
- [30] P. Tabuaciri, K.L. Bunter, H.U. Graser, Thermal imaging as a potential tool for identifying piglets at risk, AGBU Pig Genetics Workshop. Armidale, Australia:

- Animal Genetics and Breeding Unit, University of New England, 2012.
- [31] S.P. Turner, M.J. Farnworth, I.M. White, S. Brotherstone, M. Mendl, P. Knap, ... A.B. Lawrence, The accumulation of skin lesions and their use as a predictor of individual aggressiveness in pigs, Appl. Anim. Behav. Sci. 96 (3–4) (2006) 245–259.
- [32] S.P. Turner, I.M. Nevison, S. Desire, I. Camerlink, R. Roehe, S.H. Ison, Marianne Farisha, Mhairi C. Jack, R.B. D'Eath, Aggressive behaviour at regrouping is a poor predictor of chronic aggression in stable social groups, Appl. Anim. Behav. Sci. 191 (2017) 98–106.
- [33] T. Travain, E.S. Colombo, L.C. Grandi, E. Heinzl, A. Pelosi, E.P. Previde, P. Valsecchi, How good is this food? A study on dogs' emotional responses to a potentially pleasant event using infrared thermography, Physiol. Behav. 159 (2016) 80–87.
- [34] M. Valera, E. Bartolomé, M.J. Sánchez, A. Molina, N. Cook, A.L. Schaefer, Changes in eye temperature and stress assessment in horses during show jumping competitions, J. Equine Vet. Sci. 32 (12) (2012) 827–830.
- [35] M. Vollmer, K.P. Möllmann, Infrared Thermal Imaging: Fundamentals, Research and Applications, John Wiley & Sons, Wiley VCH Verlag GmbH & co, KGaA, Weinheim, Germany, 2017 (ISBN 9783527693320).