ORIGINAL ARTICLE

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Evaluation of body temperature by infrared thermography in calves under different thermal states

Avaliação da temperatura corpórea por termografia infravermelha em bezerros sob diferentes estados térmicos

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ABSTRACT: The first 30 days of calves' life is a critical period for dairy farming due to the incidence of diseases that can result in high rates of mortality and morbidity. Fever is an early clinical sign of illness; however, the use of a rectal thermometer to measure body temperature in large herds is impractical. Research using a thermographic camera has demonstrated the availability of obtaining the superficial temperature without contact the evaluated local. The objective of this study was to evaluate the use of infrared thermography to measure body temperature in calves ≤ 30 days old, identifying the body regions that present a high correlation between superficial infrared temperature (IRT) and rectal temperature. Nineteen Holstein calves were enrolled in this study. An infrared thermography camera was used to obtain thermographic images of the following body regions: hooves, eyes, forehead, muzzle, and mouth. Thermographic images were obtained at 3, 5, 10±2, 15±2 , 20±2, 25±2, and 30±2 days after birth. Maximum (Tmax), minimum (Tmin), and mean (Tmean) temperatures were obtained for each body region. Calves were categorized as hyperthermic or euthermic animals based on rectal temperature. Correlations between IRT and rectal temperature varied according to each body region and rectal temperature category. The maximum temperatures of left and right eyes measured by infrared thermography showed a higher correlation with rectal temperature, regardless of rectal temperature category (hyperthermia or euthermia). In general, Tmax and Tmean had a higher correlation with rectal temperatures than Tmin.

KEYWORDS: cattle; rectal temperature; eye; hyperthermia; euthermic.

RESUMO: Os primeiros 30 dias de vida dos bezerros são críticos na pecuária leiteira devido à ocorrência de doenças que resultam em altas taxas de mortalidade e morbidade. A febre é uma manifestação clínica precoce de enfermidade, entretanto, o uso do termômetro retal para aferir a temperatura corporal, em grandes rebanhos, é inexequível. Pesquisas demonstram a viabilidade da câmera termográfica para obter a temperatura superficial sem contato com o local avaliado. Esse estudo objetivou o uso da termografia infravermelha para mensurar a temperatura corporal em bezerros com até 30 dias de vida, a partir da identificação das regiões corpóreas que apresentam maior correlação entre a temperatura infravermelha superficial (TIV) e a temperatura retal. Dezenove bezerros Holandeses foram incluídos neste estudo. Utilizou-se uma câmera termográfica infravermelha para obter imagens das seguintes regiões corporais: cascos, olhos, fronte, muflo e boca. Obteve-se imagens aos 3, 5, 10±2, 15±2, 20±2, 25±2 e 30±2 dias após o nascimento. As temperaturas máxima (Tmax), mínima (Tmin) e média (Tmean) foram obtidas para cada região corporal. Os bezerros foram categorizados de acordo com a temperatura retal, em hipertérmicos ou eutérmicos. As correlações entre a TIV e a temperatura retal variaram de acordo com cada região corporal e categoria de temperatura retal. As temperaturas máximas dos olhos esquerdo e direito medidas por termografia infravermelha apresentaram maior correlação com a temperatura retal, independentemente da categoria do animal quanto a temperatura retal (hipertermia ou eutermia). Em geral, Tmax e Tmean tiveram maior correlação com as temperaturas retais do que Tmin.

PALAVRAS-CHAVE: bovinos; temperatura retal; olhos; hipertermia; eutérmico.

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INTRODUCTION

The first 30 days of calves' life is a critical period for dairy farming due to the incidence of diseases that can result in high rates of mortality and morbidity. In Brazil, the main causes of illness during this period are diarrheas, umbilical disorders, bronchopneumonia, sepsis, and bovine babesiosis and anaplasmosis (Feitosa *et al.*, 2001; Smith, 2006).

Fever is an adaptive response to infections and inflammatory and traumatic process. It is an early clinical sign of illness. Body temperature variations also are correlated with heat adaptation and tolerance and are an important indicator of welfare, mainly in intensive rearing systems (McManus *et al.*, 2009; McManus *et al.*, 2011).

Body temperature measurement in bovines is commonly performed using a rectal thermometer. However, physical restraint can cause stress, negatively affecting the accuracy of temperature measurement. Furthermore, using a rectal thermometer for early identification of illnesses in large herds is impractical (Schaefer *et al.*, 2012; McManus *et al.*, 2016).

The development and installation of an automated body temperature measurement system in calf barns can help farmers and managers to early identify animals with high body temperature that are more likely to progress to illness. Infrared thermography is an alternative diagnostic method, based on body temperature measurement, for early detection of illnesses and thermal stress in production animals (Schaefer *et al.*, 2004; Stewart *et al.*, 2007; Stewart *et al.*, 2008; Paim *et al.*, 2012; Schaefer *et al.*, 2012; Valera *et al.*, 2012; Costa *et al.*, 2015). Additionally, it is a non-invasive, fast, and practical method for remote temperature monitoring without the need for contact with the body surface (Speakmen; Ward, 1998; McManus *et al.*, 2016). This method allows for the identification of small increases in body temperature due to changes in blood flow under stressful situations, illnesses, and inflammatory conditions (Nääs; Garcia; Caldara, 2014; Roberto *et al.*, 2014; McManus *et al.*, 2016). The use of infrared thermography in production animals requires further studies, especially regarding image capture and evaluation in the device's software.

In this context, the objective of this study was to evaluated the use of infrared thermography to measure body temperature in calves ≤ 30 days old, identifying the body regions with a high correlation between superficial infrared temperature (IRT) and rectal temperature.

MATERIAL AND METHODS

This study was approved by Ethics Committee for Animal Use (protocol 2285150415) of the Faculty of Veterinary Medicine and Animal Science of the University of Sao Paulo (FMVZ/USP).

Animal Selection and Sampling

Convenience sampling was performed on calves housed in the facilities of the Bovine and Small Ruminant Clinic at the Faculty of Veterinary Medicine and Animal Science of the University of Sao Paulo. Male Holstein calves under 30 days old and clinically healthy (heart rate 72 to 100 beats/min, respiratory rate 20 to 40 breaths/min, pink and moist mucous membranes, non evident superficial lymph nodes and normal skin turgor; Divers; Peek, 2007) were eligible to participate in the study. Calves ≤ 3 days old and hypothermic (rectal temperature less than 37.5 °C) were excluded from the study. Animals under 3 days old exhibit significant variations in body temperature and only reach stable thermoregulation after this period of extrauterine life.

Thermographic Images

An infrared thermography camera (resolution 640×480 pixels; accuracy ± 2 °C; model T620 25°; FLIR SYSTEMS AB, Sweden) was used to obtain thermographic images. The following parameters were fixed: emissivity of 0.95; ambient temperature of 23 °C and relative air humidity of 50%; minimum distance to the camera focus varying from 0.5 to 1.0 meters; Rainbow HC color palette.

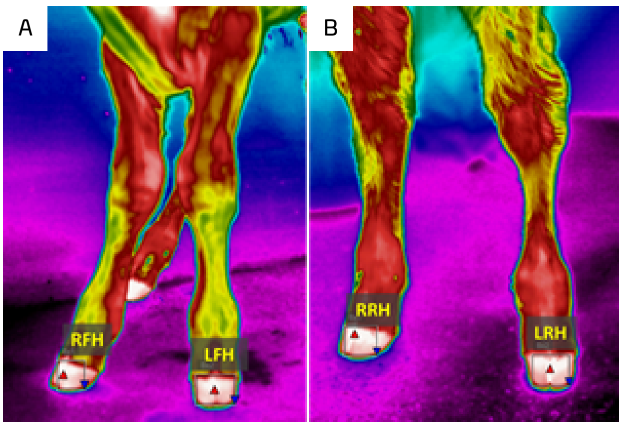
The animals were kept in enclosed barns throughout the experimental period to minimize potential effects of ambient temperature and air humidity on the thermographic images (absence of solar radiation and wind flow) (Mogg; Pollitt, 1992; Silva; Sousa Jr, 2013). During night and early morning after feeding period, calves were kept in individual barns and after that, were kept in collective barns after thermographic images were obteined. The values of these environmental parameters were fixed in the infrared thermography camera system for evaluation of the thermographic images.

Potential interferences in the thermograms, such as presence of manure or skin moisture, were avoided by keeping the calves standing in a clean and dry area and protected from direct solar radiation for at least 15 minutes before obtaining the thermographic images. No cleaning was performed on the skin surface of the photographed body region, as it would be impractical under field conditions. Thermograms were obtained during the morning period 2h after feeding to minimize the daily circadian effects.

Body Regions

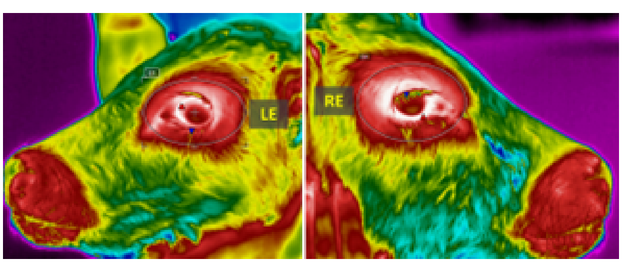
The selection of the following body regions for this study was based on previous studies (Mogg; Pollitt, 1992; Schaefer *et al.*, 2004; Schaefer *et al.*, 2007; Gloster *et al.*, 2011): right and left front hooves (RFH and LFH, respectively) and right and left hind hooves (RHH and LHH, respectively) (Figure 1), with hooves in dorsal view; right and left eyes (RE and LE, respectively) (Figure 2), with the eyeball and periocular region in lateral view; forehead (F), corresponding to the region between the eyes, including the eyeballs in rostral view; muzzle (Mz), in rostral view; and mouth (Mo), corresponding to the region near the lip commissure and muzzle in rostral view (Figure 3).

Thermographic images were obtained at 3 (D3), 5 (D5), 10±2 (D10), 15±2 (D15), 20±2 (D20), 25±2 (D25) and 30±2 (D30) days after birth. Maximum (Tmax), minimum (Tmin), and mean (Tmean) temperatures were obtained for each selected body region. A digital thermometer was used to measure rectal temperature.



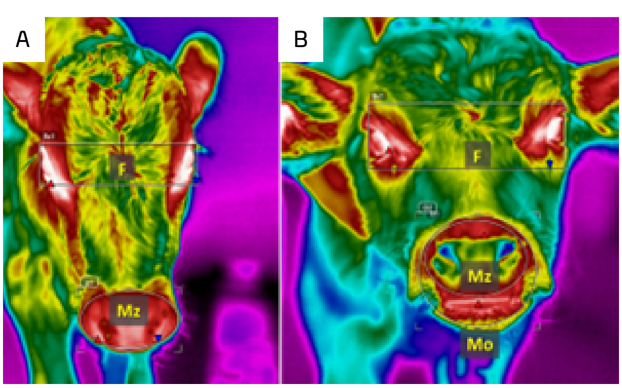
Rectangular geometric tool used to delimit the hoof areas. Right front hoof (RFH), left front hoof (LFH), right hind hoof (RRH), and left hind hoof (LRH). Arrows indicate maximum (red arrow) and minimum (blue arrow) temperatures.
Source: author's collection.

Figure 1. Thermogram of the front (A) and hind (B) limbs in cranial view.



Elliptical geometric tool used to delimit the areas. Right eye (RE) and left eye (LE). Arrows indicate maximum (red arrow) and minimum (blue arrow) temperatures.
Source: author's collection.

Figure 2. Thermogram of the head in lateral view.



Rectangular and elliptical geometric tools used to delimit the areas. Forehead (F), muzzle (Mz), and mouth (Mo). Arrows indicate maximum (red arrow) and minimum (blue arrow) temperatures.
Source: author's collection.

Figure 3. Thermogram of the head in low (A) and high (B) rostral view.

Thermographic Analysis

The thermograms were analyzed using specific software (FLIR tools®). A specific analytical geometric shape (rectangular or elliptical) was established for each body region (hooves, eyes, forehead, muzzle, and mouth) and then a specific area was delimited; from this area, the software calculated Tmax, Tmin, and Tmean in degrees Celsius (°C).

Each analytical geometric shape was selected considering the best fit for the area of interest, avoiding the inclusion of unwanted areas (such as the floor) and maximizing the intended area. Metzner *et al.* (2014) used a similar approach and achieved better results using the manual analytical tool for identifying cows with mastitis. This approach maximizes the number of evaluated pixels and reduces the interference of artifacts in thermogram analysis.

Statistical Analysis

The evaluated calves were categorized into 2 groups based on rectal temperature: hyperthermic animals, with rectal temperature ≥ 39.5 °C (RT≥39.5 °C group); and euthermic animals, with rectal temperature between 37.5 and 39.5 °C (RT<39.5 °C group).

Initially, the distribution of body surface temperature data was evaluated using graphs and the Shapiro-Wilk test for each body region, and outliers were excluded from the analysis. Pearson and Spearman correlation coefficients were used to assess the correlation between each surface temperature and rectal temperature for normal and non-normal distributed variables, respectively. Considering the normally distributed variables, generalized linear models were constructed to estimate rectal temperature as a function of body surface temperature. Coefficient of determination was used to evaluate the fit of the statistical model. Statistical analyses were performed using SAS 9.3 (SAS Institute, 2011) at a significance level of 0.05.

RESULTS

Animals and Thermographic Images

Nineteen calves met the inclusion and exclusion criteria for the study. The minimum hoof temperatures were excluded from the analysis because they coincide with the ground temperature or other regions unrelated to animal's body surface. The minimum mouth temperatures were also excluded because they included the region of the lips, which had high moisture due to the animal's salivation.

Correlation Between Superficial Infrared Temperature (IRT) and Rectal Temperature

The correlation between IRT measured by infrared thermography and body temperature measured by rectal thermometer varied according to each body region and rectal temperature category (euthermic or hyperthermic animals) (Table 1 and Figure 4).

The mouth IRT in euthermic calves showed a higher correlation with rectal temperatures, followed by muzzle IRT and forehead IRT. The left eye temperature in euthermic and hyperthermic animals had higher correlation with rectal temperature than right eye temperature. In general, Tmax and Tmean had a higher correlation with rectal temperature than Tmin (Table 1).

Regression Analysis

Following the correlation analysis, linear regression equations were constructed for each body region temperature to predict rectal temperature using the IRT of euthermic and hyperthermic calves (Table 2).

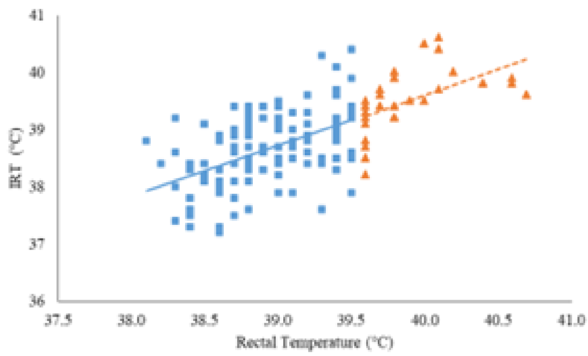
DISCUSSION

Most of the IRT measured in the body regions of euthermic calves had significant correlation ($P < 0.01$) with rectal temperature. Considering the hyperthermic calves, only Tmax of right and left eyes were significantly correlated with rectal temperature (Table 1). This difference is shown in Figure 4 (A), where the trend line (dotted line) of hyperthermic animals suggests a disproportionate increase in rectal temperature and muzzle IRT. An increase in blood flow to the skin surface may occur as a thermoregulatory response of the body to higher internal temperatures (rectal temperature), distributing and

Table 1. Pearson and Spearman correlation coefficients between infrared temperature and rectal temperature in hyperthermic and euthermic calves in each body region.

Body region infrared temperature[1]	Euthermic[2]		Hyperthermic[3]	
	Coefficient[4]	P-value[5]	Coefficient[4]	P-value[5]
Forehead				
Tmax	0.37	< 0.001	0.39	ns
Tmean	0.38	< 0.001	0.23	ns
Tmin	0.40	< 0.001	- 0.16	ns
Muzzle				
Tmax	0.46	< 0.001	0.37	ns
Tmean	0.51	< 0.001	0.33	ns
Tmin	0.43	< 0.001	0.14	ns
Mouth				
Tmax	0.52	< 0.001	0.48	ns
Tmean	0.60	< 0.001	0.44	ns
Right Eye				
Tmax	0.40	< 0.001	0.44	< 0.05
Tmean	0.36	< 0.001	- 0.06	ns
Tmin	0.25	< 0.01	0.1	ns
Left Eye				
Tmax	0.44	< 0.001	0.53	< 0.05
Tmean	0.41	< 0.001	0.01	ns
Tmin	0.26	< 0.01	0.08	ns
Left Front Hoof				
Tmax	0.35	< 0.001	0.01	ns
Tmean	0.37	< 0.001	- 0.01	ns
Right Front Hoof				
Tmax	0.68	< 0.001	- 0.14	ns
Tmean	0.46	< 0.001	- 0.14	ns
Left Hind Hoof				
Tmax	0.2	ns	- 0.24	ns
Tmean[4]	0.31	< 0.001	- 0.06	ns
Right Hind Hoof				
Tmax	0.15	ns	0.06	ns
Tmean	0.19	ns	- 0.02	ns

[1]Tmax = maximum temperature; Tmean = mean temperature; Tmin = minimum temperature.
[2]Euthermic animals, with rectal temperature between 37.5 and 39.5 °C.
[3]Hyperthermic animals, with rectal temperature ≥ 39.5 °C.
[4]The Spearman correlation test was used only for the variable Tmean of the left hind hoof in Euthermic and Hyperthermic groups (non-normal distributed variables), whereas the Pearson correlation test was used for the others variables (normal distributed variables).
[5]ns = not significant.



Squares and triangles represent euthermic and hyperthermic animals, respectively. Solid and dotted lines represent trends in euthermic and hyperthermic groups, respectively. A – Graph of maximum temperature of the muzzle. B – Graph of maximum temperature of the left eye. Source: author's collection.

Figure 4. Graphical representation of the distribution of superficial infrared temperature (IRT) data by categories of rectal temperature (euthermic and hyperthermic) in calves.

dissipating heat (Hammel; Pierce, 1968). Under hyperthermic conditions, body surface temperature is more affected by ambient temperature than rectal temperature. Mogg and Pollitt (1992) reported that during pyrexia, limb surface temperature changes disproportionately and that the interpretation should consider the ambient temperature. Gloster *et al.* (2011) found similar results and suggested that the hoof temperature is directly affected by ambient temperature.

In the present study, only Tmax of right and left eyes were significantly correlated with rectal temperature, regardless of rectal temperature category (euthermia or hyperthermia) (Figure 4). Similar results have been reported for ruminants. Dunbar *et al.* (2009) found no significant difference between eye IRT and body temperature in deer experimentally infected with foot-and-mouth disease. Gloster *et al.* (2011) reported that eye temperature was not affected by ambient temperature. Schaefer *et al.* (2004) found that the eye orbital region

Table 2. Linear regression equations for rectal temperatures predicted by infrared temperature measured by infrared thermography.

Body region infrared temperature[1]	Intercept	Coefficient	R^2 [2]	P-value[3]
Forehead				
Tmax	25.8	0.346	0.292	< 0.01
Tmean	36.3	0.076	0.147	< 0.01
Tmin	37.6	0.043	0.160	< 0.01
Muzzle				
Tmax	32.3	0.185	0.307	< 0.01
Tmean	35.0	0.118	0.312	< 0.01
Tmin	36.9	0.073	0.230	< 0.01
Mouth				
Tmax	31.3	0.207	0.230	< 0.01
Tmean	33.8	0.152	0.397	< 0.01
Right Eye				
Tmax	23.2	0.408	0.290	< 0.01
Tmean	31.4	0.209	0.182	< 0.01
Tmin	36.5	0.077	0.108	< 0.01
Left Eye				
Tmax	21.3	0.458	0.373	< 0.01
Tmean	32.0	0.195	0.178	< 0.01
Tmin	32.0	0.068	0.075	< 0.01
Left front Hoof				
Tmax	36.7	0.063	0.123	< 0.01
Tmean	37.2	0.053	0.139	< 0.01
Right front Hoof				
Tmax	36.6	0.065	0.135	< 0.01
Tmean	36.7	0.066	0.210	< 0.01
Left Hind Hoof				
Tmax	36.5	0.071	0.095	< 0.01
Tmean	-	-	-	ns[4]
Right Hind Hoof				
Tmax	36.8	0.062	0.062	< 0.01
Tmean	37.4	0.049	0.077	< 0.01

[1]Tmax = maximum temperature; Tmean = mean temperature; Tmin = minimum temperature.
[2]Coefficient of determination
[3]P-value of the regression model
[5]ns = not significant.

was more sensitive to changes in body temperature than muzzle, ears, or back regions in animals inoculated with bovine viral diarrhea. Additionally, Schaefer *et al.* (2007) reported that measuring IRT in eyes is a method for early diagnosis of bovine respiratory diseases.

Hind hoof temperatures (Tmax and Tmean of LHH and RHH) had no significant correlation with rectal temperature for both euthermic and hyperthermic calves. However, the front hoof temperatures were significantly correlated with rectal temperature in euthermic calves. This difference may be due to the asymmetry of the thermographic images of the hind hooves (angular images); thus, the rectangular geometric shapes used during thermogram evaluation were smaller, reducing the number of measured pixels and increasing artifact interference (Metzner *et al.*, 2014).

Overall, the coefficient of determination (R^2) from the linear regression analysis was relatively low for all investigated body regions. Despite the measures taken to avoid potential environmental effects on the thermographic images (animals kept standing in closed barns and protected from direct sunlight for 15 minutes prior to image capture), the circadian variation in skin surface temperature may have affected the results (Berry *et al.*, 2003; Scarpellini; Bícego, 2010).

Tmax of left and right eyes presented the highest correlation with rectal temperature, presenting the highest R^2 in the linear regression analysis, and were independently correlated with rectal temperature categories (euthermia or hyperthermia). This result is consistent with previous studies that reported that measuring IRT in eyes is a potential method for assessing body temperature in bovines (Schaefer *et al.*, 2004; Schaefer *et al.*, 2007; Stewart *et al.*, 2008; Dunbar *et al.*, 2009; Gloster *et al.*, 2011).

The authors believe that results of the present study can be extrapolated to a larger population of similar calves to those included in this study. Our findings can be used to develop an automated system for measuring body temperature based on infrared thermography, assisting farmers and managers in the early detection of illnesses and thermal stress in calves. In this context, eyes may be a potential body region to achieve this purpose through IRT measurement. Thus, further studies should be conducted to investigate risk factors associated with measurement of body surface temperature in calves using infrared thermography.

CONCLUSIONS

The maximum temperatures of left and right eyes measured by infrared thermography showed a higher correlation with rectal temperature in Holstein newborn calves, regardless of rectal temperature category (euthermia or hyperthermia). Additionally, the correlation between superficial infrared temperature and body temperature varied according to each body region and rectal temperature category (euthermic and hyperthermic animals). The findings of this study can be used to develop an automated system for measuring body temperature based on infrared thermography, assisting farmers and managers in the early detection of illnesses and thermal stress in calves.

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