# Effects of infrared camera angle and distance on measurement and reproducibility of thermographically determined temperatures of the distolateral aspects of the forelimbs in horses

Simone Westermann, Dr med vet; Heinz H. F. Buchner, Dr med vet, PhD; Johannes P. Schramel, Dr med vet; Alexander Tichy, Dr rer nat; Christian Stanek, Prof Dr med vet

**Objective**—To assess effects of camera angle and distance on measurement and reproducibility of thermographically determined temperatures of the distolateral aspect of the forelimbs in horses.

Design—Evaluation study.

Animals—10 adult horses.

**Procedures**—Thermographic images of both forelimbs were obtained at 3 times during the day (replicates 1, 2, and 3); maximum surface temperature over 1 region (distolateral aspect of the third metacarpal bone and metacarpophalangeal joint) was measured. Standard images were obtained every 5 minutes for 1 hour with the camera positioned at an angle of 90° and a distance of 1.0 m from the forelimb; additional images were obtained at changed ( $\pm$  20°) angles or at a 1.5-m distance. At the end of each replicate, 4 sets of additional images were obtained at 2-minute intervals to assess short-term reproducibility.

**Results**—Mean  $\pm$  SD temperature difference between left and right forelimbs was 0.32°  $\pm$  0.27°C (0.58°  $\pm$  0.49°F) in standard images. Temperatures measured via standard images were highly correlated with those measured with the camera positioned at changed angles or distance. Mean  $\pm$  SD differences between temperatures measured via standard images and those measured from changed angles or distance were considered small ( $\leq$  0.22°  $\pm$  0.18°C [0.40°  $\pm$  0.32°F] for all comparisons). The degree of short-term reproducibility was high.

**Conclusions and Clinical Relevance**—Thermographically determined temperatures were unaffected by 20° changes in camera angle or a 0.5-m increase in camera distance from the forelimb. Minor temperature differences between left and right forelimbs were detected in the study and should be considered during diagnostic investigations. (*J Am Vet Med Assoc* 2013;242:388–395)

Infrared thermography has been used as a diagnostic tool in veterinary medicine since the mid-1960s.<sup>1-3</sup> Particularly in the field of equine orthopedics, several studies have been published in which this technology was used.<sup>2,4,5</sup> The procedure is noninvasive, the equipment is easy to handle, and results are provided rapidly with minimal or no risk of injury to the animal and investigator.<sup>6</sup> The camera detects infrared radiation (heat) emitted by the body surface and produces an image in which the color gradient corresponds with the distribution of surface temperatures. Thermographic imaging may be useful for detecting differences in the expected distribution patterns of temperature among or within limbs and identifying areas of inflammation or vascular stenosis that may potentially be associated with lameness.<sup>2,5–7</sup> Generally, normal thermal patterns are established by vessel topography and anatomic structures. At the distal aspects of the limbs in horses, the third metacarpal and metatarsal bones, metacarpophalangeal and metatarsophalangeal (fetlock) regions, and proximal interphalangeal (pastern) joint typically have cool temperature patterns relative to the coronary band. Between the flexor tendons and the third metacarpal or metatarsal bone, a strip of increased temperature attributable to presence of the digital palmar artery and vein is detectable.<sup>1,6</sup>

The accuracy of thermography is still under discussion because ambient temperature and other immediate environmental conditions, such as radiation of the sun or airflow, may influence surface temperatures<sup>2,5,6,8,9</sup> in regions of interest and may hamper the interpretation of images. The authors of 2 previous studies<sup>1,2</sup> recommended performing thermography at an ambient temperature of approximately 20°C (68°F) or at temperatures < 30°C (< 86°F) for thermographic evaluation in horses. It has also been recommended that horses should be kept in the examination room for 10 to 20 minutes prior to ther-

From the Units of Large Animal Surgery and Orthopaedics (Westermann, Buchner, Stanek) and Internal Medicine (Schramel), Equine Clinic, and Department of Biomedical Sciences (Tichy), University of Veterinary Medicine Vienna, Veterinärplatz 1, A-1210 Vienna, Austria.

The authors thank Andreas Angerer at InfraTec GmbH for technical support.

Address correspondence to Dr. Westermann (Simone.Westermann@ vetmeduni.ac.at).

mography to allow for temperature equilibration.<sup>1,2,8</sup> However, few studies have been performed to assess the influence of immediate environmental conditions on thermography<sup>10–13,a</sup> and the reproducibility of thermographic findings.<sup>6,14,15</sup> In clinical practice, time constraints and patient movement can result in deviations of camera angle and distance from the camera to the region of interest when obtaining thermographic images for diagnostic purposes. These variables in particular may vary among examinations and could potentially affect the results of thermographic imaging.

The objective of the study reported here was to assess the effects of changes in camera angle and distance between the camera and region of interest (ie, imaging distance) on measurement and reproducibility of thermographically determined temperatures of the distolateral aspects of the forelimbs of horses. We hypothesized that a 20° increase or decrease in camera angle and a change in imaging distance from 1.0 to 1.5 m would not significantly affect thermographically determined temperatures or the reproducibility of thermographic measurements in this region. Additionally, we measured ambient environmental temperatures and relative humidity and rectal temperature of the horses to assess correlation of these variables with imaging results.

#### **Material and Methods**

Animals—Ten adult mixed-breed (6 warmblood-type and 4 trotter-type) horses owned by the Equine Clinic of the University of Veterinary Medicine, Vienna, were used in the study. The 6 geldings and 4 mares ranged from 9 to 22 years of age, with body weights of 480 to 654 kg (1,056 to 1,439 lb). All horses were determined to be free of lameness at walk and had no clinical signs of acute illness or injuries. Horses were housed in a paddock full-time, and each had a short, fine haircoat considered appropriate for the summer season. The experimental protocol was approved by the institutional Animal Welfare and Ethics Committee of the University of Veterinary Medicine, Vienna.

**Experimental procedures**—Thermographic imaging was performed with a portable infrared camera<sup>b</sup> equipped with a 12.5-mm focal length lens, an uncooled microbolometer, and a focal plane array infrared detector with a spectral range between 7.5 and 14  $\mu$ m. Emissivity was adjusted to 1.00 (unitless value). A bubble level was fixed on the camera to assure horizontal orientation.

All measurements were performed on 1 day for each horse. The evening before experiments were performed, the horses were stabled in a box stall and groomed. The next day, they were brought to the examination room (requiring an approx 200-m walk outdoors) and placed in stocks. The room was approximately 40 m<sup>2</sup> in size; all doors and windows were kept closed, and air-conditioning was switched off. After 1 set (1 replicate) of measurements was completed, the horses were brought back to the box stall until the next replicate was performed. Thermographic imaging was performed at 3 times during the day, starting at 8:00 AM (replicate 1), 12:00 noon (replicate 2), and 4:00 PM (replicate 3). Prior to each replicate of thermographic measurements, the ambient temperature and relative humidity<sup>c</sup> of the stable (in which horses were kept on the day of the experiments) and of the immediate outdoor environment were recorded. Ambient temperature and relative humidity in the examination room and rectal temperature<sup>d</sup> of the horses were recorded prior to each replicate of measurements and 30 minutes and 1 hour after the replicate was started.

Angle and distance were determined with a wooden caliper that included a 20° angle and had a total length of 1.5 m, with a mark 1.0 m from the end placed nearest to the horse. To evaluate equilibration time, thermographic images of the distolateral



Figure 1—Photograph of the forelimbs of a horse standing in stocks for thermographic imaging of the distolateral aspect of the left forelimb. The infrared camera is placed at a 90° angle to the plane of the region of interest and a distance of 1.0 m to obtain a standard image. The wooden caliper used for distance and angle measurements is shown. The short arm of the caliper is offset 20° in a dorsolateral direction from the standard imaging angle (ie, 70°).



Figure 2—Thermographic image of the region of interest for temperature measurement (including the lateral aspect of the third metacarpal bone and metacarpophalangeal joint) in the right forelimb of a horse. The white line marks borders of the region (indicated as X11 in the image). The maximum temperature for the region is indicated in the lower box. Temperature values were color coded as indicated by the scale bar on the right.

aspects of the right and left forelimbs were recorded at 5 minute-intervals over a period of 60 minutes. Standard images were obtained at a camera height of approximately 15 cm and a distance of 1.0 m with an angle of 90° (total number of measurements in each replicate, 13/limb). At the beginning of each replicate (time 0) and after 30 and 60 minutes, additional images were recorded at angles of 70° and 110° with a distance of 1.0 m, as well as at 90° with a distance of 1.5 m, for both forelimbs. Starting with the left forelimb each time, the angle of imaging was changed in a dorsolateral direction  $(70^\circ)$ , and that of the right forelimb was changed in a palmarolateral direction  $(110^\circ)$ , for a total of 3 images/limb at each angle in a replicate (**Figure 1**). Images at a distance of 1.5 m were obtained for each forelimb, for a total of 3 images/limb in each replicate.

Short-term reproducibility was also tested at the end of each replicate. A series of images (1 standard image/limb plus 1 image at each additional angle

Table 1—Mean, minimum, and maximum temperatures of the regions of interest (distolateral aspect of the left and right forelimbs, including the third metacarpal bone and metacarpophalangeal joint) measured in standard thermographic images of healthy horses obtained during 3 measurement replicates performed at 8:00 AM (replicate 1), 12:00 noon (replicate 2), and 4:00 PM (replicate 3).

Replicate		Na af	Temperature (°C)			
horses)	Forelimb	measurements	Mean ± SD	Minimum	Maximum	
1 (10)						
. (,	Left	165	$32.5 \pm 1.1$	29.5	34.1	
	Right	162	$\textbf{32.6} \pm \textbf{1.2}$	29.6	34.5	
2 (10)						
	Left	154	$\textbf{32.9} \pm \textbf{0.8}$	31.1	34.6	
0 (0)	Right	156	$\textbf{33.0} \pm \textbf{0.8}$	31.4	34.4	
3 (8)	Loft	101	22 1 J 0 E	22.1	21.2	
	Right	120	$33.1 \pm 0.5$ $33.2 \pm 0.5$	32.1	34.3 34.3	

Standard images were obtained with the infrared camera placed at a 90° angle to the plane of the region of interest and a distance of 1.0 m. Images were obtained at 5-minute intervals for 1 hour in each replicate; 4 additional images were obtained at 2-minute intervals at the end of each replicate. Measurements were not obtained from 2 horses in replicate 3 because of technical problems and because horses were uncooperative. In each replicate, a small number of results was excluded from analysis because of technical problems. To convert temperature from Celsius to Fahrenheit, multiply by 9/5 and add 32.





Figure 3—Box-and-whiskers plots of temperatures determined in standard thermographic images of the distolateral aspects of the left and right forelimbs (dark gray and light gray bars, respectively) in healthy horses. Images were obtained every 5 minutes for 1 hour in each of 3 replicates performed at 8:00 AM (replicate 1; n = 10 [A]), 12:00 noon (replicate 2; 10 [B]), and 4:00 PM (replicate 3; 8 [C]). A series of additional images was obtained at 2-minute intervals to evaluate short-term reproducibility at the end of each replicate. Measurements were not obtained from 2 horses in replicate 3 because of technical problems and because horses were uncooperative. In each replicate, a small number of results was excluded from analysis because of technical problems. Upper and lower limits of each box represent the 25th to 75th percentiles, and horizontal lines within boxes indicate the median values. Whiskers indicate values up to 1.5 times the interquartile range of the lower and upper quartiles, respectively. To convert temperature from Celsius to Fahrenheit, multiply by 9/5 and add 32.

[70° and 110°] and 1 image/limb at 90° with a 1.5-m distance) was obtained at 2-minute intervals within an 8-minute period. These were compared with the last measurements of the previous replicate (ie, the 60-minute values). Differences were further calculated between successive temperature recordings (first vs second, second vs third, and third vs fourth) by use of these values.

Images were analyzed with commercially available software.<sup>e</sup> Maximum surface temperature was calculated from a region of interest which consisted of a polygon overlying the distolateral aspect of the third metacarpal bone and fetlock joint (**Figure 2**). Absolute values and changes in temperatures among standard images and images at additional angles and distance were determined.

Statistical analysis—A trigonometry formula was used to determine that an angle change of 20° is equivalent to a variation of 35 cm in the horizontal camera position (with a fixed distance of 1.0 m between camera and forelimb). Image data were recorded and analyzed with statistical software.<sup>f</sup> Because data were normally distributed (Kolmogorov-Smirnov test), mean temperatures between left and right forelimbs and between standard images of the left or right forelimb and those obtained at changed angles or distance were compared with a paired t test. Pearson correlation analysis was used to determine linear correlation of mean temperatures between the left and right forelimbs and between forelimb temperatures and ambient temperatures, relative humidity, and rectal temperatures of horses. The development of temperatures within each measurement replicate was analyzed with a general linear model to compare the differences between temperatures (contrasts) over time. Differences were compared by means of repeated-measures ANOVA. Values of  $\tilde{P} < 0.05$  were accepted as significant.

### Results

Standard images—Measurements were obtained from all 10 horses for the first 2 replicates and from

8 horses for replicate 3. Measurements were not obtained from 2 horses in replicate 3 because of technical problems and because horses were uncooperative. In each replicate, some images were excluded from analysis because of technical problems, typically related to image quality. Mean temperatures of the region of interest for each forelimb in standard images (camera angle, 90°; distance, 1.0 m) were evaluated for the 3 measurement replicates (Table 1). The greatest difference between the lowest and the highest temperatures for a single horse within 1 limb was 1.62°С (2.92°F) in replicate 1 (8:00 AM), 1.56°С (2.81°F) in replicate 2 (12:00 noon), and 1.37°C (2.47°F) in replicate 3 (4:00 PM). The variation of the temperature values was apparently greater within replicate 1 than in replicate 3, although this was not evaluated statistically.

Temperature profiles over time for the region of interest in the left and right forelimbs in each of the 3 replicates (determined for standard images) were summarized with box-and-whiskers plots (Figure 3). Although mean temperature values appeared to reach a plateau after approximately 40 minutes in replicate 1, this could not be confirmed with statistical methods. With a general linear model that compared the

Table 3—Within-replicate correlation (r) between temperatures determined by use of standard thermographic images and images obtained with the camera positioned at a changed angle or increased distance relative to the standard image for the same horses in Table 1.

	Replicate			
Comparison	1	2	3	
Left vs right forelimb (standard image) Standard image vs changed angle	0.92	0.90	0.77	
Left forelimb (70°)	0.98	0.97	0.95	
Right forelimb (110°) Standard image vs increased distance	0.93	0.94	0.94	
Left forelimb	0.97	0.96	0.91	
Right forelimb	0.97	0.95	0.92	
All correlations were significant ( <i>P</i> < <i>See</i> Tables 1 and 2 for remainder of k	0.001). ey.			

Table 2—Mean, minimum, and maximum temperatures determined by use of standard thermographic images and images obtained with the camera positioned at a changed angle or increased distance relative to the standard image for the same horses in Table 1.

	Standard Image		Changed angle		Increased distance (1.5 m)	
Variable	Left forelimb	Right forelimb	70° (left forelimb)	110° (right forelimb)	Left forelimb	Right forelimb
No. of measurements Temperature (°C)	440	438	178	172	169	170
Mean ± SD Minimum Maximum	$\begin{array}{r} 32.8 \pm 0.9 \\ 29.5 \\ 34.6 \end{array}$	$\begin{array}{r} 32.9 \pm 0.9 \\ 29.6 \\ 34.5 \end{array}$	$\begin{array}{r} 32.7 \pm 0.9 \\ 29.3 \\ 34.5 \end{array}$	$\begin{array}{c} 32.9 \pm 0.8 \\ 29.9 \\ 34.3 \end{array}$	$\begin{array}{c} 32.7 \pm 0.9 \\ 29.2 \\ 34.4 \end{array}$	$\begin{array}{r} 32.7 \pm 0.9 \\ 29.8 \\ 34.4 \end{array}$

For images obtained at changed angles, the camera was positioned at the same distance as for the standard image; the angle of imaging for the left forelimb was changed in a dorsolateral direction (70°), and that of the right forelimb was changed in a palmarolateral direction (110°). For images obtained at the 1.5-m distance, the camera was positioned at the same angle as for the standard image (90°). Images at changed angle and increased distance were obtained at the beginning of each replicate and at 30 and 60 minutes; 4 additional images of each type were obtained at 2-minute intervals at the end of each replicate.

See Table 1 for remainder of key.

differences between temperatures (contrasts) over time, no significant difference could be found between any of the values. Temperatures measured via standard images of the left forelimb were positively correlated with those of the right forelimb (r = 0.90; P < 0.001).

Changes in angle and distance of the camera—Temperatures measured by use of standard images of the left and right forelimb and images obtained at changed angles (70° and 110°) or distance (1.5 m at 90°) in the 3 measurement replicates were summarized (**Table 2**). Strong positive correlations between the temperatures of the left and right forelimb were found, independent of changes in angle (r = 0.91; P < 0.001) or distance (r = 0.91; P = 0.012).

Temperatures measured via standard images of the left forelimb were positively correlated with those obtained when the camera was positioned at different angles and at a greater distance (r = 0.98 and 0.96, respectively). A similar degree of correlation was also found between the same measurements for the right forelimb (r = 0.90 and 0.90, respectively). All correlations between thermographic temperatures for a given limb within a replicate were significant (P < 0.001) at all camera positions tested (**Table 3**).

Mean difference in temperature between regions of interest in the left and right forelimbs as measured via standard images was evaluated (**Table 4**). Mean temperature differences for each forelimb when the camera was positioned at the 70° or 110° angle or at the 1.5-m distance were then compared with the temperature difference measured in standard images of the respective limb. These values were not significantly different. When compared among the 3 replicates, most of the differences appeared to decrease after replicate 1, and the differences in temperature change between standard images and those obtained at different angles were significantly (P < 0.001 for both forelimbs) smaller for replicate 3, compared with replicate 1 (**Table 5**).

Associations with ambient temperature, relative humidity, and rectal temperature—Mean ambient temperature and relative humidity of the stable where horses were kept, the outdoor environment,

and the examination room, as well as the rectal temperature of horses were assessed for each replicate (Table 6). During the days when measurements were performed, mean  $\pm$  SD outdoor temperature increased from  $24.1^{\circ} \pm 2.5^{\circ}C$  (75.38° ±  $4.50^{\circ}F$ ) to  $30.8^{\circ} \pm 3.3^{\circ}C (87.44^{\circ} \pm 5.94^{\circ}F)$  and that in the examination room increased from  $25.9^{\circ} \pm 1.7^{\circ}C$  (78.62)  $\pm 3.06^{\circ}$ F) to 27.8°  $\pm 1.3^{\circ}$ C (82.04°  $\pm 2.34^{\circ}$ F; these differences were not evaluated statistically). All of the ambient temperatures measured and relative humidity in the examination room were positively and significantly (P < 0.025) correlated with temperatures measured via thermography, regardless of the distance (1.0 or 1.5 m) or angle (70°, 90°, or 110°) at which the camera was placed (Table 7). Similar correlations were not detected with relative humidity in the stable or outdoor environment or with rectal temperature.

Short-term reproducibility—Reproducibility experiments at the end of the replicates revealed a mean temperature difference in thermographically determined temperatures of  $0.16^{\circ} \pm 0.14^{\circ}$ C (0.29  $\pm 0.25^{\circ}$ F) for the left forelimb and  $0.19^{\circ} \pm 0.15^{\circ}$ C (0.34°  $\pm 0.27^{\circ}$ F) for the right forelimb. Differences between the highest and lowest temperature for a single horse in standard images were  $0.33^{\circ}$ C (0.59°F) for the left forelimb and  $0.38^{\circ}$ C (0.68°F) for the right forelimb. This numeric difference between the forelimbs was not significant.

Table 5—Mean within-replicate temperature differences (°C) for standard thermographic images and images obtained with the camera positioned at a changed angle or increased distance relative to the standard image for the same horses in Table 1.

	Replicate		
Comparison	1	2	3
Left vs right forelimb (standard image) Standard image vs changed angle	0.36	0.30	0.31
Left forelimb (70°)	0.22ª	0.16 <sup>b</sup>	0.14 <sup>b</sup>
Right forelimb (110°)	0.21ª	0.18	0.13 <sup>⊾</sup>
Standard image vs increased distance (1.5 m)			
Left forelimb	0.25	0.18	0.20
Right forelimb	0.25	0.21	0.19
<sup>a,b</sup> Within a row, values with different super- nificantly ( <i>P</i> < 0.001) different. <i>See</i> Tables 1 and 2 for remainder of key.	script l	etters a	re sig-

Table 4—Differences between temperatures determined by use of standard thermographic images and images obtained with the camera positioned at a changed angle or increased distance relative to the standard image for the same horses in Table 1.

	N. (	Temperature difference (°C)			
Comparison	No. of measurements	$\textbf{Mean} \pm \textbf{SD}$	Minimum	Maximun	
Left vs right forelimb (standard image) Standard image vs changed angle	413	$\textbf{0.32} \pm \textbf{0.27}$	0.00	1.32	
Left forelimb (70°)	176	$\textbf{0.18} \pm \textbf{0.15}$	0.00	0.90	
Right forelimb (110°) Standard image vs increased distance (1.5 m)	166	$\textbf{0.18} \pm \textbf{0.17}$	0.00	1.07	
Left forelimb	165	$\textbf{0.21} \pm \textbf{0.19}$	0.00	1.08	
Right forelimb	164	$\textbf{0.22} \pm \textbf{0.18}$	0.00	0.98	

Table 6—Mean  $\pm$  SD ambient temperatures, relative humidity, and rectal temperatures of the same horses in Table 1 within replicates 1, 2, and 3.

	No. of	Replicate			
Variable	no. of measurements	1	2	3	
Ambient temperature (°C)					
Outdoors	27	$24.1 \pm 2.5$	$\textbf{27.3} \pm \textbf{2.4}$	$\textbf{30.8} \pm \textbf{3.3}$	
Stable	26	$\textbf{23.4} \pm \textbf{2.1}$	$\textbf{26.2} \pm \textbf{1.5}$	$28.1 \pm 1.5$	
Examination room	82	$\textbf{25.9} \pm \textbf{1.7}$	$\textbf{26.8} \pm \textbf{1.4}$	$\textbf{27.8} \pm \textbf{1.3}$	
Relative humidity (%)					
Outdoors	27	$52.3\pm4.0$	$\textbf{45.0} \pm \textbf{8.8}$	$\textbf{38.5} \pm \textbf{9.5}$	
Stable	26	$53.7\pm4.9$	$\textbf{49.9} \pm \textbf{7.1}$	$44.7\pm8.1$	
Examination room	82	$50.9\pm4.2$	$\textbf{47.9} \pm \textbf{7.0}$	$46.4\pm7.6$	
Rectal temperature of horses (°C)	83	$\textbf{37.6} \pm \textbf{0.2}$	$\textbf{37.6} \pm \textbf{0.3}$	$\textbf{37.7} \pm \textbf{0.3}$	

Ambient temperature and relative humidity of the stable and of the immediate outdoor environment were recorded prior to each replicate. Ambient temperature and relative humidity in the examination room and rectal temperature of the horses were recorded prior to each replicate and 30 minutes and 1 hour after the replicate was started.

*See* Table 1 for remainder of key.

Table 7—Correlation (r) of environmental variables and rectal temperature of horses with thermographic temperatures measured from various angles and distances for the same horses in Table 1.

	Standard image		Changed angle		Increased distance (1.5 m)	
Variable	Left forelimb	Right forelimb	70° (left forelimb)	110° (right forelimb)	Left forelimb	Right forelimb
Ambient temperature (°C)						
Outdoors	0.80*	0.71*	0.81*	0.75*	0.79*	0.76*
Stable	0.90*	0.90*	0.75*	0.81*	0.90*	0.90*
Examination room	0.84*	0.86*	0.85*	0.87*	0.82*	0.89*
Ambient relative humidity (%)						
Outdoors	0.13	0.03	0.10	0.05	0.08	0.00
Stable	0.01	0.04	0.02	0.04	0.06	0.02
Examination room	0.26*	0.30*	0.26*	0.29*	0.27*	0.35*
Rectal temperature	0.01	0.02	0.03	0.02	0.02	0.04

## Discussion

Several reports<sup>1,2,4,7,16-19</sup> describing the use of thermographic imaging have been published, particularly in the field of orthopedics. To ensure that the information obtained is comparable among different thermographic images, these should be standardized as much as possible by placement of the camera at the same angle and the same distance each time.14,20 Time constraints and movement of patients may cause problems that result in unwanted changes in camera angle and distance within an imaging session. Moreover, the angle and distance of the camera may vary among repeated measurements and reexaminations. Therefore, we assessed the extent to which the thermographically determined temperature was affected by 20° changes in angle or a 0.5-m increase in distance between the camera and the region of interest at the distolateral aspect of the forelimbs. These values were chosen to simulate situations in practice where changes in camera position are made inadvertently. In addition, we analyzed imaging results during a 1-hour equilibration period, among repeated measurements within a short-term (8-minute) period after equilibration, and between 3 measurement replicates performed at different times during the day (8:00 AM [replicate 1], 12:00 noon [replicate 2], and 4:00 PM [replicate 3]). We also analyzed associations of ambient temperature and relative humidity with these results.

In the present study, temperatures of the region of interest in the left and right forelimbs measured via standard thermographic images (camera angle, 90°; distance, 1.0 m from the distolateral aspect of the forelimb) were closely correlated within each of 3 measurement replicates and among all 3 replicates. Consistent with our results, a high degree of symmetry of the thermographic pattern between the left and right third metacarpal and third metatarsal bones has been described.<sup>6,12,15</sup> Some authors have indicated that a difference of  $\geq 1^{\circ}$ C (1.8°F) in thermographically determined temperatures between corresponding areas in contralateral limbs is clinically relevant and may indicate pathological processes.<sup>1,5,8</sup> The magnitude of temperature variations caused by differences in methods must to be considered to avoid inaccuracies in interpreting results. In our study, the mean  $\pm$  SD difference between temperatures in standard images of the left and right forelimbs was  $0.32^{\circ} \pm 0.27^{\circ}C (0.58^{\circ} \pm 0.49^{\circ}F)$ . A temperature deviation of 0.59°C (1.06°F; including SD) is within the specification of the infrared camera used in the present study and can be recommended as a threshold for detection of thermographic abnormalities in the described region. However, it is important to always evaluate thermographically determined temperature differences in combination with a detailed anamnesis and clinical examination.<sup>7</sup>

The influence of changes in camera position on thermographic images of the dorsal aspect of the right and left hands of humans has been described.14 In that study, 14 investigators each created 2 sequential thermographic images of the dorsal aspect of both hands of 2 patients. When positioning of the hands was changed slightly, individual errors of measurement for different regions up to  $2.3^{\circ} \pm 0.54^{\circ}$ C ( $4.14^{\circ} \pm 0.97^{\circ}$ F) for the left hand and up to  $1.2^{\circ} \pm 0.49^{\circ}C (2.16^{\circ} \pm 0.88^{\circ}F)$  for the right hand were found.<sup>14</sup> The authors of that study hypothesized that the differences may have been caused by a high degree of variability in temperatures of the hands between subjects and that the repeatability of standardized thermographic views varies among different anatomic regions. It remained unclear whether the demonstrated effects were influenced by individual differences in vasculature of the hands. However, they concluded that the positioning of standardized views are reproducible in a narrow range by different investigators.14,g

In the present study, the greatest differences between thermographic temperatures determined with standard images and with images obtained at the same distance from angles of 70° (left forelimb; dorsolateral direction) and 110° (right forelimb; palmerolateral) were 0.90° and 1.07°C (1.62° and 1.93°F) for the left and right forelimbs, respectively, with mean differences of 0.18°C (0.32°F) for both limbs. This indicates only a small and nonsignificant effect attributable to changes of 20° in angle of the camera position.

Trigonometry calculations revealed that an angle change of 20° is equivalent to a variation of 35 cm in horizontal camera position (with a fixed distance of 1.0 m between the camera and forelimb). Results of the present study indicate that an investigator can deviate 35 cm horizontally from the 90° angle at a 1.0-m distance from the limb without significantly influencing the thermographically determined temperature.

It has also been described that a change in infrared camera distance from a subject influences the results because of technical effects.<sup>20</sup> A thermographic image taken from a greater distance results in a smaller region of interest with fewer pixels and lower resolution.<sup>20</sup> This effect can be minimized by use of uncooled microbolometer focal plane arrays as was done in the present study. With this technique, changes that result in a smaller region of interest should have no effect on image quality and resolution.<sup>21</sup> Despite a high degree of correlation between these variables, mean temperature differences between standard images and images obtained at the 1.5-m distance were 0.21°C (0.38°F) for the left forelimb and 0.22°C (0.40°F) for the right forelimb. The maximum difference was 1.08°C (1.94°F). Changes in distance from 1.0 to 1.5 m had no significant effect on recorded temperatures.

We can speculate whether the results from our study are transferable to other anatomic regions in horses. Because the distal limb of the horse has several large blood vessels directly under the skin, individual patterns of vasculature may affect thermographically determined temperatures, and even in this region, changes in camera position had only small effects on thermographically determined temperature. Therefore, we assume that in other regions, changes in the angle and distance of the camera would not substantially affect the determined temperature. This should be properly evaluated in further studies.

In the study reported here, we analyzed thermographic temperature variations within a 60-minute equilibration period at various times during the day. Some authors have recommended that horses should remain in the examination room for 10 to 20 minutes before imaging to allow for temperature equilibration.<sup>1</sup> One group reported that only 19% of horses reached a thermographically determined temperature plateau (measured regions: lateral metacarpophalangeal joint, lateral thorax, and gluteal regions) after equilibration of 10 to 20 minutes, whereas 25% reached plateau values after 21 to 38 minutes, and 56% required > 39 minutes.<sup>15</sup> Mean time to equilibration was 39 minutes and was influenced by environmental temperature. Unfortunately, the authors did not report exact ambient temperature values. During the first replicate in our study, temperatures of the right and left forelimbs appeared to reach a plateau after approximately 40 minutes. However, it was not possible to prove this statistically. In replicate 2, only a weak increase to a visibly detectable plateau was apparent, and in replicate 3, values had the smallest variations over time and no apparent plateau was detected. It can be speculated that this lack of equilibration was caused by minor increases in ambient outdoor temperatures, which had a mean value of approximately 30.8°C during this replicate. Analyses of thermographically determined temperatures among the 3 replicates revealed that variations among temperatures decreased from replicate 1 to replicate 3. It remains unclear whether this was caused by diurnal variations in the horses or by adaptations to ambient temperature. One explanation could be that the blood vessels were fully vasodilated with increasing ambient temperatures approaching values closer to body core temperature and remained vasodilated during the day.<sup>22</sup>

In the present study, thermographically determined temperatures of the left and right forelimbs were positively correlated with ambient temperatures, which is in agreement with results of other studies<sup>12,23</sup> in horses and ponies. Authors of 1 study<sup>23</sup> ascertained a time-dependent temperature equilibration of the forelimbs in ponies, especially when the ambient temperature was between 18° and 20°C (64.4° and 68°F). Those authors also proposed contralateral forelimb temperature differences (left vs right) of about 1.5°C (2.7°F) as representing normal biological variation or a subclinical inflammatory process, rather than indicating pathological changes. They concluded that obtaining thermographic images of the patient in a room with an ambient temperature > 20°C should facilitate interpretation of temperature differences between forelimbs.<sup>23</sup> In our study, all ambient temperatures remained > 20°C throughout the study period. The hypothesis that ambient temperature has an effect on equilibration<sup>23</sup> was supported by the temperature changes among replicates in our study, which decreased from replicates 1 to 3 as the ambient temperatures increased.

To evaluate short-term reproducibility of the measurements in our study, a series of images (1 standard image/limb plus 1 image at each additional angle [70° and 110°] and 1 image/limb at 90° with a 1.5-m distance) was obtained at 2-minute intervals within an 8-minute period, and temperature values were compared among these time points and with the final measurements of the previous replicate. Mean temperature differences of 0.16° and 0.19°C (0.29° and 0.34°F) for the left and right forelimbs, respectively, in standard images demonstrated a high degree of reproducibility. The authors of other studies<sup>1,6,15</sup> reported that circulatory patterns and blood flow in the limbs of horses influence the use of thermal patterns as a basis for interpretation. They found that the general characteristics of thermal patterns did not change during thermographic imaging and that no 2 horses have exactly the same pattern.<sup>1,6,16</sup> In the present study, we could subjectively detect differences to support this perception, but it was not possible to prove it graphically or statistically. To confirm variations in the individual thermographic patterns of the distal forelimbs of horses, further investigations are required.

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