



A Comparative Evaluation of the Alternative Anatomical Sites for Body Temperature Measurement Using Digital Thermometers in Dairy Cows

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ABSTRACT

The measurement of body temperature is a critical aspect of assessing the health and reproductive status of dairy cows. The standard method used to estimate this temperature is rectal thermometry. However, this technique has limitations, including disease spread, distress, and or risks of rectal injuries. The current study was undertaken to validate the potential of alternative anatomical sites for temperature measurement using a digital thermometer (DT). The study employed a one-factor experimental design considering the anatomical site as the main factor, with four treatments or factor levels, namely rectal (DT_{rectal}), inguinal (DT_{inguinal}), axillary (DT_{axillary}), and undertail (DT_{undertail}) sites. A simple random sampling technique was employed to determine the order of site selection for temperature measurement. In total, 26 adult Holstein Friesian-Boran cows with an average weight of 482 kg were used to conduct this study. Each cow was assessed for all the treatments considered in this study. The temperature measured at different anatomical sites was evaluated. The highest mean temperature was observed for rectal temperature (38.27 ± 0.42°C), while that of mean axillary temperature was the lowest (37.75 ± 0.53°C). The mean temperature readings were significantly affected by the anatomical site. There was no significant difference between mean rectal and inguinal or undertail temperature. There was a significant correlation between the rectal and undertail temperature, while no significant correlation was observed between rectal and inguinal temperature. The equivalence analysis between the rectal and undertail pair revealed a significant bias. This bias suggests that the two anatomical sites cannot be used interchangeably, particularly with digital thermometer application in Holstein Friesian-Boran cows. However, the observed mean undertail temperature and its correlation with rectal temperature indicated that the undertail site still holds promise as an alternative site for temperature-taking under conditions different from this study.

Keywords: Anatomical site, Dairy cow, Digital thermometer, Temperature

INTRODUCTION

Cattle productivity and production in sub-Saharan Africa, including Zambia, have generally stagnated despite the increasing demand for animal protein (Omollo et al., 2020). The low productivity and production have been attributed to diseases, inadequate veterinary services, climate change effects, and poor husbandry practices (MFL, 2020; Odubote, 2022). There have been calls for appropriate measures to address reproductive and productive insufficiency in cattle (MFL, 2020; Sianangama et al., 2022). One of the crucial measures is the early diagnosis of any physiological or pathological condition that may influence cattle performance (Godyń et al., 2018). Additionally, core body temperature is among the parameters considered during the clinical examination of these animals. In this case, it serves as a crucial physiological marker for the cows' health and reproductive status (Debnath et al., 2017). Examples of health and reproductive conditions whose diagnosis may be facilitated by temperature detection include infectious diseases, thermal stress, estrus synchronization, estrus status, and the onset of calving (Fischer-Tenhagen and Arlt, 2020). Routine temperature detection enables timeous decision-making or management of such conditions, thereby minimizing undue reproductive and economic losses (Godyń et al., 2018; Abigaba and Sianangama, 2023).

Measurement of body temperature in cattle has seen the development of various means, including clinical mercury, clinical digital, non-contact infrared thermometers, thermal infrared cameras, and temperature loggers (Pourjafar et al., 2012; Sellier et al., 2014). While these devices hold significance, a majority of them come with drawbacks such as high cost, complexity, potential hazards, reduced accuracy, or limited availability (Muhammed et al., 2019; Marquez et al., 2021). These limitations discourage clinicians or farmers from using these devices on animals, particularly in developing countries. Many smallholder farmers in Zambia have poor husbandry skills (Odubote, 2022). These farmers lack

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adequate veterinary services, especially in rural areas, and they fail to detect estrus, pregnancy, thermal stress, or even take the temperatures of their cows, which downgrades the usefulness of many thermometer types. According to Tagesu (2018), farmers play a crucial role during the clinical examination of animals by providing information during history-taking by a clinician or veterinarian. If the farmers could take or monitor their animals' temperature, clinicians would timeously diagnose many conditions. Notably, any changes in the core body temperature can be considered an early threat to cow's health (Fischer-Tenhagen and Arlt, 2020). Hence, there is an urgent need to further explore the reliability of temperature devices that are cheap and easy to use, particularly by farmers who spend more time with their animals than clinicians.

Rectal thermometry is the gold standard for body temperature assessment in cows (Zubor et al., 2020; Giannetto et al., 2022). However, this method is associated with potential risks, such as stress, disease spread, and rectal injuries to cattle (Yadav et al., 2017). Temperature taking in cows per rectum is conventionally performed using a mercury thermometer (MT) or digital thermometer (DT). Although MT is reliably accurate, it is slow in detecting rectal temperature. Additionally, there is a concern that this device could potentially introduce mercury contamination into the environment or pose a risk of injury to the cow if the thermometer breaks. On the contrary, the DT is generally safer, readily available, user-friendly, with acceptable accuracy, and relatively rapid (Cadioli et al., 2010; Hine et al., 2015). These advantageous qualities have led to a preference for the DT over MT.

It is noteworthy that the DT device may also serve as a fomite for disease transmission or cause rectal injury and distress to the cows, particularly when it is applied per rectum (Muhammed et al., 2019). Kearton et al. (2020) confirmed that measuring body temperature at the peripheral locations may be helpful and less invasive to an animal. This is particularly important in cases where clinicians and/or farmers cannot measure the temperature per rectum in local cows (genotypes) with a retractable temperament (Muhammed et al., 2019). Previous studies have confirmed the accuracy and reliability of some anatomical sites for body temperature measurement using DT or MT. Examples include the armpit in humans (Chaturvedi et al., 2004) and the inguinal site in chickens (Abigaba and Sianangama, 2023). However, little is known about the potential of various skin locations for temperature measurement in cows using contact-DTs. Given the aforementioned limitations, there exists a need to search for non-invasive, safer, and user-friendly anatomical sites for body temperature measurement in cows. It is preferred that any identified anatomical site should be robust to external variations given that the conventional measurement approach has shown associations with variations in rectal temperature (Ramey and Lee, 2011; Pourjafar et al., 2012). This study was carried out to explore the reliability of less-risky anatomical sites for the body temperature assessment in dairy cows using a DT.

MATERIALS AND METHODS

Ethical approval

The study was approved by the Institutional Committee on Animal Research, University of Zambia, Lusaka, Zambia (No. 1595-2021). The procedures used for this study were non-lethal to the study animals. Animal handling, including feeding and watering, restraint, and experimentation, was performed with strict supervision by the institutional committee on animal research. These were done in compliance with the guide for the care and use of agricultural animals in research and teaching (ASAS, 2020).

Study area

This study was conducted at the Field Station, Department of Animal Science, University of Zambia, Lusaka, Zambia. The research was carried out during the month of February-March 2023. Zambia lies in the tropics, within the Southern African region. The GeoNames geographical database Google Earth-2023 located her at latitude S 14° 20' 0" and longitude E 28° 30' 0". During the study period, the average ambient temperature and relative humidity at the field station ranged from 23.4 to 31.7°C and 50 to 79%, respectively.

Experimental animals

This study included the Boran-Holstein Friesian crossbred cows that belonged to the Department of Animal Science, University of Zambia, Lusaka, Zambia. These were physically healthy dairy cows with different parities and unconfirmed status of gravidness. The weight of these cows ranged from 314 to 680 kg, with an average of 482 kg. The cows were within the age range of 4-8 years, with an average of 6.5 years.

Study design

This research employed a single-factor experimental design to determine the effect of anatomical site on the body temperature estimation in dairy cows. According to the study design, the anatomical site was the main factor that had four levels, including rectal, inguinal, axillary, and undertail sites. The temperature measurement for each site (DT_t) constituted a treatment; hence, four treatments, namely rectal temperature (DT_{t_{rectal}}), inguinal temperature (DT_{t_{inguinal}}),

axillary temperature ($DT_{axillary}$), and undertail temperature ($DT_{undertail}$) were considered for this study. A total of 26 cows were used for this study, and each animal was assessed for DT_{rectal} , $DT_{inguinal}$, $DT_{axillary}$, and $DT_{undertail}$. Additionally, 26 temperature measurements were performed for each anatomical site, with the DT_{rectal} considered as the control. Before the temperature measurement, each cow was physically restrained according to the procedures introduced by Tagesu (2018). The DT readings were taken from each cow while in a chute (restrained) after more than 15 minutes of lapse. The 15-minute lapse was intended to minimize the potential effects of psychogenic fever on the study results.

The DT measurements were conducted using a functional veterinary digital thermometer (DT; GB Kruise digital thermometer, Taipei, Taiwan). This device had a measuring range of 30-43.9°C and a resolution of 0.1°C. The order of the sites to be measured was determined using a simple random selection. Therefore, folded papers bearing the name of each site were tossed, followed by picking one of them randomly without replacing it. This procedure was repeated twice for each study cow. Subsequently, the temperatures, including DT_{rectal} , $DT_{inguinal}$, $DT_{axillary}$, and $DT_{undertail}$ readings, were obtained. The rectal temperature (DT_{rectal}) was measured following a previous procedure put forward by Pourjafar (2012). In the case of the axillary and inguinal methods, the procedure for the measurement of DT was based on an earlier study by Levy et al. (2020). Briefly, the $DT_{axillary}$ was measured by inserting a DT probe deep into the left axilla, approaching from the caudal aspect, and aiming towards the dorsum. The measurement procedure was conducted on a cow standing with its forelimb close to the body. Similarly, the $DT_{inguinal}$ readings were obtained from the cows in this posture. To obtain the $DT_{inguinal}$ readings, the DT probe was inserted deep in the left inguinal area, approaching from the cranial aspect, and aiming towards the dorsum. On the other hand, the undertail temperature was obtained by introducing a DT probe in between the ano-triangular surface and tail base, approaching from the lateral aspect, and aiming towards the cranial direction. For each site, the device was left in position (about 15-50 seconds) until a degree sign stopped flashing and an alarm went off. Additionally, the DT readings for each site were taken twice, and their average was considered a single datum. Regarding the potential effects of ambient temperature on the DT readings, all measurements were performed during the morning hours (8:30-11:30 a.m.).

Data analysis

The data on the DT readings were analyzed in the Statistical Package for Social Scientists (SPSS® IBM 26 version, USA) using selected descriptive and inferential statistics. The normality and homogeneity of the data were checked using Shapiro-Wilk and Lavene's tests, respectively. The descriptive statistics included means and standard deviations, while the inferential statistics were correlation, ANOVA, and equivalence tests. The F-test in ANOVA (One-way) was used to determine the main effect of anatomical site on the DT readings. The following statistical model was used

$$Y_{ij} = \mu + \beta_i + e_{ij}$$

Where, Y_{ij} is the dependent variable representing the DT reading, μ indicates the overall mean, β_i denotes the main effect of the site factor with four levels (i = rectal, inguinal, undertail, and axillary sites), and e_{ij} is the error term. The Least Significance Difference (LSD) statistic was employed to ascertain the treatments/pairs who's mean DT readings differed significantly. The correlation between the different anatomical sites (DT readings) was determined using a Pearson's correlation test. The one-sample t-test was employed to establish the level of bias between each alternative anatomical site and the control (rectal thermometry). In all the tests, significance was taken at $p < 0.05$.

RESULTS

The mean temperature readings at different anatomical sites of adult Holstein Friesian-Boran cows

The mean temperature readings (DT), including DT_{rectal} , $DT_{inguinal}$, $DT_{axillary}$, and $DT_{undertail}$ were obtained from the rectal, inguinal, axillary, and undertail sites, respectively (Table 1). The highest mean value was observed for the DT_{rectal} ($38.27 \pm 0.42^\circ\text{C}$), while the $DT_{axillary}$ had the lowest mean value ($37.75 \pm 0.53^\circ\text{C}$). The smallest difference in mean temperature values was observed between DT_{rectal} (control) and $DT_{undertail}$ (0.19°C). There was a significant effect of anatomical site on the mean DT readings (F [3, 100] = 7.08, $p < 0.05$, $\eta_p^2 = 0.175$). In this case, 17.5% of the variability in temperature readings between the different treatments was explained by this factor (anatomical site). The mean DT_{rectal} was significantly different from that of the $DT_{axillary}$ ($p < 0.05$). There was no statistical difference in mean values of DT_{rectal} and $DT_{undertail}$ ($p > 0.05$).

Correlation between temperature readings at different anatomical sites of adult Holstein Friesian-Boran cows

The results from correlation analysis (bivariate) of the DT readings, including DT_{rectal} , $DT_{inguinal}$, $DT_{axillary}$, and $DT_{undertail}$, are presented in Table 2. The correlation between the DT_{rectal} and $DT_{undertail}$ readings was significantly stronger ($r = 0.889$, $p < 0.05$) than other pairs. The results revealed a weak correlation between DT_{rectal} and $DT_{inguinal}$ readings ($r = 0.102$, $p > 0.05$). Similarly, a weak correlation between $DT_{inguinal}$ and $DT_{undertail}$ ($r = 0.154$, $p > 0.05$) was observed.

Reliability of selected temperature measurements at different anatomical sites of adult Holstein Friesian-Boran cows

The results of reliability analysis that quantitatively analyzed the statistical significance between the paired DT readings are presented in Table 3. The largest mean of differences (bias) was observed between DT_{axillary} and DT_{rectal} readings ($-0.53 \pm 0.43^{\circ}\text{C}$), while the DT_{undertail} and DT_{rectal} pair had the lowest bias ($-0.19 \pm 0.19^{\circ}\text{C}$). The t-test (one-sample) on the mean of differences for the DT_{undertail} and DT_{rectal} pair revealed a significant bias ($p < 0.05$). There was a significant bias between the DT_{rectal} and DT_{inguinal} pair ($p < 0.05$), and a similar finding was revealed for the DT_{axillary} and DT_{rectal} pair ($p < 0.05$).

Table 1. The mean temperature readings at different anatomical sites of adult Holstein Friesian-Boran cows

Anatomical site	DTt readings (°C)	Mean \pm SD (°C)	Difference from DT _{t_{cloacal}} (°C)	Minimum (°C)	Maximum (°C)
Rectal		38.27 ± 0.42^a	-	37.20	39.05
Inguinal		38.06 ± 0.33^a	0.21	37.15	38.55
Axillary		37.74 ± 0.53^b	0.53	36.70	38.75
Undertail		38.08 ± 0.38^a	0.19	37.25	38.70

DTt: Temperature reading by a digital thermometer, SD: Standard deviation, °C: Degrees Celsius, ^{ab}Different superscript letters within the same column indicate a significant difference ($p < 0.05$).

Table 2. Correlation between temperature readings at different anatomical sites of Holstein Friesian-Boran cows

	DT _{t_{rectal}}	DT _{t_{inguinal}}	DT _{t_{axillary}}	DT _{t_{undertail}}
DT _{t_{rectal}}	1			
DT _{t_{inguinal}}	0.102	1		
DT _{t_{axillary}}	0.601**	0.324	1	
DT _{t_{undertail}}	0.889**	0.154	0.596**	1

DTt: Temperature readings by a digital thermometer, correlation coefficient 0.00-0.10: negligible, 0.10-0.39: Weak, 0.4-0.69: Moderate, 0.7-0.89: Strong, 0.9-1.0: Very strong correlation, **significant correlation at $p < 0.05$

Table 3. Reliability of selected measurement methods at different anatomical sites of adult Holstein Friesian-Boran cows

Paired sites/methods	DTt difference (°C)	Mean \pm SD (°C)	df	T value	P value	95% CI	
						Lower	Upper
Undertail-Rectal		-0.19 ± 0.19	26	-0.508	< 0.05	-0.27	-0.11
Inguinal-Rectal		-0.21 ± 0.51	26	-2.109	< 0.05	-0.41	0.00
Axillary-Rectal		-0.53 ± 0.43	26	-6.228	< 0.05	-0.71	-0.35

DTt: Temperature readings by a digital thermometer, SD: Standard deviation, Df: degrees of freedom, CI: Confidence interval, °C: Degrees Celsius, <: Lower than

DISCUSSION

In an effort to search for anatomical sites that are non-invasive and less disease-risky, with DT application, the current study has revealed that the mean temperature at the undertail site is largely similar to that of the rectal temperature, unlike the situation observed at other anatomical sites. It was also confirmed that anatomical site had an effect on the mean temperature readings, which agreed with the previous study findings that reported an effect of the skin point on the thermometer readings (Abioja et al., 2019; Abigaba and Sianangama, 2023). Additionally, the observed mean temperature value at the undertail site was generally consistent with an established body temperature range ($38-39.3^{\circ}\text{C}$) for dairy cattle (Reece, 2009). This finding was probably attributed to the anatomical position of the tail, which closely covers the ano-triangular surface and consequently minimizes the heat loss, compared to the other skin locations. According to Abioja et al. (2019), the accuracy of the body temperature measurement depends on the type of thermometer used and the location or anatomical site. It should be noted that the observed numerical variation between the rectal and undertail temperature, along with other anatomical sites, aligns with previous findings in thermometry studies involving different species (Chaturvedi et al., 2004; Levy et al., 2020). The numerical difference could be related to the skin temperature, as it relates to external temperature, which is generally lower than the internal body temperature. Skin generally loses more heat to the environment and is metabolically less active (Abioja et al., 2019; Levy et al., 2020).

This study indicated a stronger correlation between rectal and undertail temperature, when compared to the case of rectal temperature with other thermometry methods. Consequently, the outcomes generally lend support to undertail thermometry as a promising alternative for approximating rectal temperature, especially when compared to inguinal and

axillary methods. In a previous study, where a micro-chip (temperature sensor) was inserted in the vulvar muscles, it was suggested that the vulva was an appropriate proxy for rectal temperature (Morais et al., 2006). However, the potential damage to the vulva downgrades its usefulness (Kou et al., 2017). Considering the relationship between core body temperature and temperatures at the vulva and rectum, it is reasonable to expect a strong correlation between the temperature recorded at the undertail site and rectal temperature in the current study. The findings of this study could be attributed to the anatomical disposition or orientation of ano-triangular surface in relation to the tail that snugly covers it. Moreover, the undertail site lacks thick hair, which has been associated with inaccurate readings during temperature measurement using a non-contact infrared device (Kou et al., 2017).

The inguinal temperature was not correlated with rectal (standard) temperature despite the similarity in their mean temperature values. This observation was probably attributed to the varied functional status of the mammary glands. In other words, some cows were lactating while others were generally dry. This notion is based on the previous study findings indicating a significant correlation between mammary/inguinal temperature and milk production (dos Santos et al., 2022). According to Zaborski et al. (2022), the correlation between udder skin temperature and milk presence is attributed to the increased blood flow into the udder. Moreover, the potential effect of heat generated during milk synthesis on the inguinal temperature readings cannot be underestimated. However, the temperature of lactating cows tends to lower after milking (Araki et al., 1984). An enlarged udder would further reduce the thigh-udder (inguinal) space, reducing heat loss to the environment (Golzarian et al., 2017). Considering udder enlargement, it is plausible that pregnancy status and parity of the cows were also potential sources of the observed temperature disparity. In cows, the udder enlarges during pregnancy, similarly, the volume of mammary glandular tissue differs between nulliparous and primiparous or multiparous cows (Davis, 2017; Zhao et al., 2019). Both mammary stroma and the glandular tissue increase during pregnancy, additionally, a significant lobuloalveolar structure is maintained during involution in ruminants such as cows (Zhao et al., 2019). The above-mentioned factors contribute to the inaccuracy of inguinal thermometry, which, as the case may have been in the current study, compromises the suitability of the inguinal site for body temperature assessment.

Although correlation coefficients measure the strength of a linear relationship between two variables or methods, the same test does not determine the agreement between these methods (Doğan, 2018). In the current study, the correlation between rectal and undertail temperature readings was strong, however, the results of equivalence analysis for this pair indicated a significant bias. One of the key features of performing body scoring in dairy cattle is the level of fat around the tail head, which reduces proportionally as the cow's body condition score decreases (Klopčič et al., 2011). The tail surface (base) snugly covers the ano-triangular surface in cows with over 2.5 body condition score on a 5-point scale. However, the gap (space) between the tail and ano-triangular surfaces widens with a sunken tail head in those with a poorer condition score. This discrepancy is anticipated to result in greater heat loss in the latter group compared to cows with a better body condition, which probably contributed to the observed bias. Another reason for the observed bias is the pregnancy status of cows. Kim et al. (2021) found a significant mean difference in ruminal temperature between pregnant and non-pregnant cows, which was attributed to the thermogenic effect of progesterone. A similar finding was reported for the vaginal temperature in cows (Suthar et al., 2012). Accordingly, the effect of progesterone, with or without body condition score, on the undertail temperature may be an important factor to ponder on. Regardless of the cause, the implication of this bias is that undertail and rectal thermometry methods cannot be substituted for one another when a DT is used to measure the body core temperature of Holstein Friesian-Boran cows.

Although the current study indicates that the undertail and rectal thermometry are not interchangeable, under the above conditions, it is not clear whether the same results would hold when factors such as body score, breed, age, and gender are considered. Of note, some earlier studies reported an association of factors like breed and sex with the level of disagreement between the rectal, axillary, and inguinal temperature measurements in other species, such as dogs and humans (Chaturvedi et al., 2004; Harper et al., 2023). Moreover, any breakthrough with the discovery of a suitable thermometry method, particularly using a DT, will be crucial for promoting improved performance in cattle (Rubia-Rubia et al., 2010). Furthermore, the measurement of body temperature plays a pivotal role in dairy cattle, aiding in the detection of estrus, pregnancy, calving onset, disease, inter alia, which contributes to timeous management decision-making and improved reproductive efficiency (Fischer-Tenhagen and Arlt, 2020; Szenci, 2022).

CONCLUSION

The present study has found that the undertail and rectal temperature have similar mean values, and a strong correlation, distinguishing them from the other investigated anatomical sites. However, the results of equivalence analysis revealed a significant bias between these two thermometry methods. For this reason, undertail thermometry cannot serve as a direct substitute for the conventional rectal method, particularly when a digital thermometer is applied to measure the body temperature in Holstein Friesian-Boran dairy cows. It is recommended that further studies be conducted using a larger sample size and on different breeds and age groups of cattle for generalization of the current findings.

DECLARATIONS

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Authors' contribution

Rubaijaniza Abigaba conceived, designed, collected and analyzed data, and wrote the manuscript. Pharaoh Collins Sianangama designed, supervised the study, and reviewed the manuscript. Both authors read and approved the final manuscript for publication.

Conflict of interests

The authors declare no conflict of interest regarding this publication.

Ethical consideration

The authors declare that this manuscript is original, and is not being considered elsewhere for publication. Other ethical issues including consent to publish, misconduct, fabrication of data, and redundancy have been checked by the authors.

Availability of data and materials

The additional data from the present study may be provided on request from the corresponding author.

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