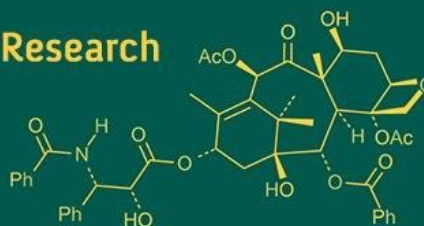


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Determination of the suitable time for infrared thermography-based tick identification

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Abstract

Accurate quantification of tick infestation is crucial for effective tick control in livestock, yet conventional visual counting methods are constrained by observer bias, hair cover, and environmental influences. Infrared thermography (IRT) offers a non-invasive alternative for tick detection based on thermal contrast between ticks and host skin; however, the optimal time for image capture under tropical conditions remains unclear. This study aimed to identify the most suitable time window for IRT-based tick identification and quantification in crossbred cattle. Five naturally tick-infested crossbred cattle (12-24 months old) maintained under uniform management conditions and without acaricide treatment were monitored. Thermal images were captured hourly from 6:00 A.M. to 12:00 noon using a high-resolution infrared camera. Tick counts were obtained by visual inspection and automated analysis of IRT images in ImageJ, with emphasis on skin and tick-surface temperature differences across body regions. Skin surface temperature was significantly higher ($p < 0.01$) than tick surface temperature in the inner thigh, perineal, and dewlap regions, with the greatest temperature difference observed in the inner thigh (2.5 ± 0.15 °C). Automated IRT analysis showed maximum tick detection between 6:00 and 10:00 A.M., followed by a decline toward mid-day as ambient temperature increased and thermal contrast decreased. The study concludes that early morning (6:00-10:00 A.M.) is the most suitable time for IRT image capture for accurate tick identification and quantification in crossbred cattle under tropical climatic conditions. IRT combined with automated image analysis represents a reliable tool for precision tick surveillance.

Keywords: Infrared thermography, ImageJ, tick identification

Introduction

Ticks are among the most economically important ectoparasites affecting livestock in tropical and subtropical regions, causing substantial losses through reduced productivity, increased disease transmission, control costs, and deterioration of hide quality (Geevarghese *et al.*, 1997; Ghosh *et al.*, 2007) [3, 4]. Crossbred cattle are particularly susceptible to tick infestation, and accurate assessment of tick burden is essential for the timely and effective implementation of control strategies (Khan *et al.*, 2013; Asmaa *et al.*, 2013; Ragulraj S *et al.*, 2023) [5, 1, 7]. However, precise quantification of ticks under field conditions remains challenging due to limitations associated with conventional visual counting methods, including small tick size, hair cover, skin pigmentation, and observer variability (Cortivo *et al.*, 2016) [2].

Infrared thermography (IRT) has emerged as a non-invasive, objective tool in precision livestock farming for detecting ectoparasites by measuring surface-temperature differences between parasites and hosts. Previous studies have demonstrated that tick surface temperature is generally lower than that of the host animal, enabling their identification through thermal contrast (Cortivo *et al.*, 2016) [2]. The effectiveness of IRT, however, is highly dependent on external factors such as ambient temperature, solar radiation, humidity, and time of image capture, all of which influence the thermal gradient between ticks and the animal's skin (Zheng *et al.*, 2022; Ghanghas *et al.*, 2025) [8, 9]. Several recent studies have emphasized that suboptimal environmental conditions and inappropriate image capture timing can reduce thermal contrast and compromise the accuracy of IRT-based detection under field conditions (Polsky *et al.*, 2022) [11].

Under tropical climatic conditions, environmental temperature increases rapidly from early morning to midday, potentially reducing the temperature difference between ticks and skin surface and thereby affecting detection accuracy. As the day progresses, increased ambient heat may elevate tick surface temperature, mask thermal contrast and make reliable identification difficult. Despite the demonstrated potential of IRT for tick detection, limited information is available on the optimal time window for image capture to maximize thermal contrast and improve counting accuracy under field conditions. Therefore, identifying the most suitable time of day for IRT image acquisition is critical to standardizing this technique for practical application in livestock tick surveillance. In this context, the present study was undertaken to determine the optimal time window for infrared thermography-based tick identification and quantification in crossbred cattle under tropical climatic conditions, using automated ImageJ analysis.

Materials and Methods

The study was conducted at the Livestock Research Centre (LRC) of the ICAR-National Dairy Research Institute (ICAR-NDRI), Karnal, Haryana, at an elevation (altitude) of 250 meters above mean sea level, latitude 29°43" North and longitude 77°20" East. This is a tropical climate region, and the annual minimum and maximum temperatures range from 4 to 45 °C. During the hot-dry season (April to June), the relative humidity varies from 41 to 85 per cent and the vapour pressure from 7.0 to 25 mmHg. Ethical approval (number 45-IAEC-19-3) was sought from the Institutional Animal Ethics Committee (IAEC) at ICAR-National Dairy Research Institute for the protocols followed and the animals' management in the experiment.

The animals were maintained under uniform management conditions. Five crossbred animals aged between 12 and 24 months infested with ticks were selected randomly. The animals were not subjected to acaricide treatment. The animals were gathered early at 5.00 A.M. in a covered, fenced area with a shed and natural tree shade in June. The same animals were monitored every hour from 6.00 A.M. to 12.00 Noon for manual tick counting and IRT imaging. The animals were restrained individually and left unattended for 2-3 minutes. Two methods were used for tick counting: visual counting and automated counting using ImageJ software based on thermal images. Ticks were manually counted by skilled veterinarians by the visual method in various regions (lateral aspect of trunk, inner thigh area, perineal region, and dewlap) of the right side of the body and then the value was multiplied by two for estimation of total tick count as described by Cortivo *et al.* (2016)^[2] with little modification in selecting the regions of the body. The second method was thermographic image analysis for auto-counting using ImageJ software. Thermographic images of all the animals were captured using a handheld, high-resolution (384 x 288 pixels) digital infrared thermal camera (Darvi DTL007, TAK Technologies Pvt. Ltd.). The thermal camera can capture temperatures from -20 °C to +650 °C. The thermal sensitivity of the IRT camera was < 20 °C with an accuracy of ±2%. Before capturing the IRT image, the camera was calibrated to ambient temperature and humidity. The reference calibration was performed using the camera cap, which was stored at ambient temperature. The emissivity and reflected apparent temperature were kept

constant at 0.96 and 20 °C, respectively, for all images, as recommended by the manufacturer for biological tissue analysis. The image was processed using thermal image analysis software (Darvi TI analysis software) provided by the manufacturer of the IRT camera.

The temperature of the tick and skin surface was measured. The software was used to convert the default colour palette (Rainbow) to "inverted grey" to improve image definition (Figure 1), and the resulting image was then used to auto-count the ticks in ImageJ (<http://imagej.nih.gov/ij/>). The values generated from visual and thermal images, coupled with automated analysis using ImageJ, were evaluated to determine the optimal time for infrared thermography.

Results and Discussion

The results presented in Figure 2 show that skin surface temperature was significantly higher ($p < 0.01$) than tick surface temperature in the inner thigh (35.30 ± 0.07 vs. 32.80 ± 0.12 °C), perineum (34.61 ± 0.31 vs. 33.24 ± 0.35 °C), and dewlap regions (36.31 ± 0.07 vs. 35.48 ± 0.13 °C). Although skin surface temperature was numerically higher than tick surface temperature on the lateral side (35.84 ± 0.12 vs. 35.10 ± 0.07 °C), the difference was not statistically significant. These findings agree with those of Cortivo *et al.* (2016)^[2], who reported lower tick surface temperatures than host skin temperatures. The observed thermal contrast between ticks and the host body surface can therefore be effectively exploited for detecting tick infestation in livestock. The relatively higher tick surface temperature in the lateral and dewlap regions may be attributed to a denser hair cover and greater exposure to ambient conditions compared to ticks in the inner thigh and perineum regions (Cortivo *et al.*, 2016)^[2].

The maximum temperature difference between tick and skin surface (2.50 ± 0.15 °C) was recorded in the inner thigh region. It was significantly higher ($p < 0.01$) than that observed in the perineum (1.37 ± 0.11 °C), dewlap (0.83 ± 0.16 °C), and lateral regions (0.74 ± 0.15 °C) (Figure 3). Cortivo *et al.* (2016)^[2] similarly reported a temperature difference of approximately 1 °C between ticks and host skin. The greater thermal contrast in the inner thigh region may be due to the absence of hair and minimal exposure to environmental temperature. In contrast, dense hair growth and increased exposure in the dewlap and lateral regions likely reduce thermal differentiation. The distinct temperature difference observed in the inner thigh region underscores its suitability for IRT image acquisition, as it minimizes intrinsic and extrinsic interference factors and enables more accurate detection and quantification through image-processing software.

Infrared thermographic images of the inner thigh region, analysed using ImageJ, revealed that the highest tick counts were observed in images captured between 6:00 and 10:00 AM, followed by a gradual decline as the day progressed (Figure 4 and Table 1). Based on these observations, the early morning period (6:00-10:00 AM) appears to be the most suitable time window for capturing IRT images for tick identification and enumeration. The decline in detection efficiency later in the day may be associated with increasing ambient temperature, which likely influences tick surface temperature and reduces the thermal contrast between ticks and host skin, thereby limiting accurate identification using IRT.

Infrared thermography has increasingly been recognized as a reliable non-invasive imaging technology in livestock research, with its effectiveness influenced by environmental conditions such as ambient temperature and solar radiation (Zheng *et al.*, 2022) ^[8]. As ambient temperature rises throughout the day, the thermal gradient between host skin and ectoparasites tends to diminish, reducing the visibility of ticks in thermal images. This pattern is consistent with broader findings from animal thermal imaging studies, which show that environmental interference significantly affects the accuracy of surface temperature measurements under field conditions (Zheng *et al.*, 2022; Ghanghas *et al.*, 2025) ^[8, 9]. Therefore, optimal image acquisition for tick identification requires selecting periods with relatively stable, lower ambient temperatures to maximise thermal contrast.

Recent advancements in infrared thermography applications in livestock highlight the importance of timing and methodological optimization to improve diagnostic performance. These include integrating artificial intelligence and enhanced imaging protocols to reduce noise from environmental and physiological variables, thereby improving temporal consistency and reliability of thermal patterns (Kim *et al.*, 2025). Within this context, the early morning window (6:00-10:00 AM) consistently emerged as the most suitable period for capturing high-quality thermograms for tick identification, due to greater thermal differentiation and reduced environmental interference. Thus, early morning IRT imaging represents an optimal approach for reliable tick detection using infrared thermography under tropical field conditions.

Table 1: Tick count variation analysed by Image J in inner thigh region in different hours during forenoon period

Time	Number of ticks in Inner thigh				
	Animal No. 782	Animal No. 796	Animal No. 773	Animal No. 782	Animal No. 997
6 AM	32	16	48	14	16
7 AM	32	22	40	16	16
8 AM	32	22	40	12	15
9 AM	32	7	32	18	16
10 AM	28	9	32	17	13
11 AM	22	9	27	10	13
12 AM	23	14	28	10	14

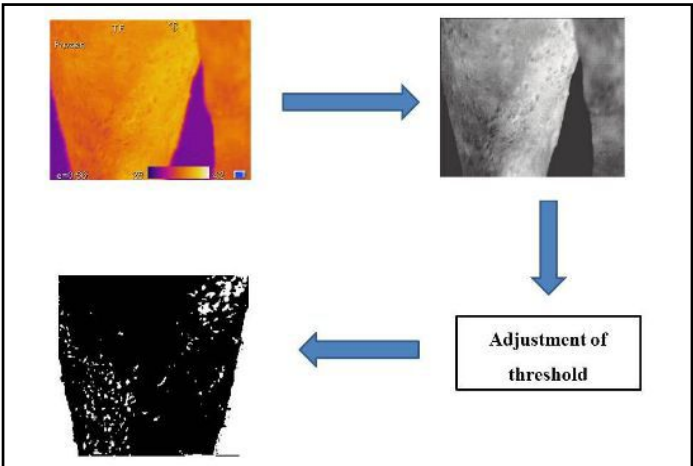


Fig 1: Tick count through IRT images and Image J analysed images.

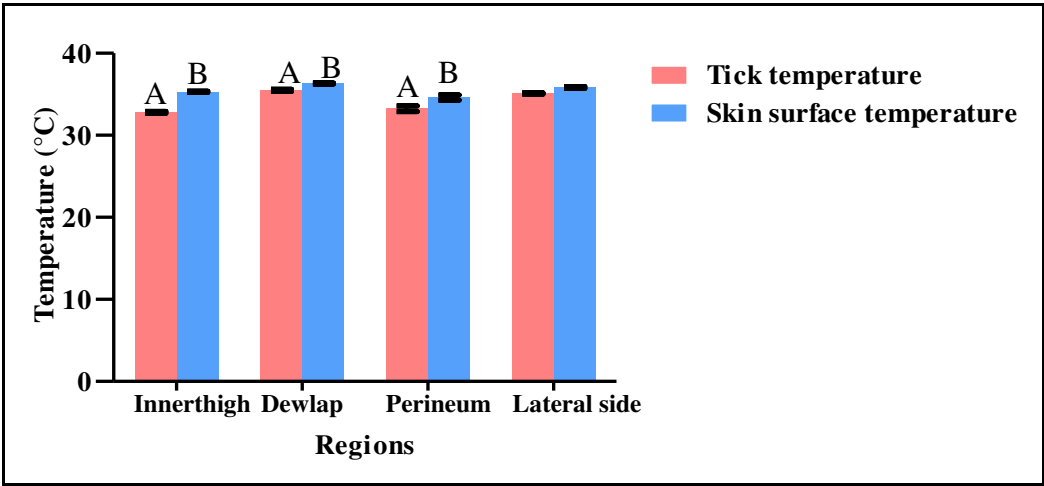


Fig 2: Tick and skin surface temperature (°C) in various regions of animal body and IRT images; Bars with different superscripts differ significantly ^{AB} ($p<0.01$)

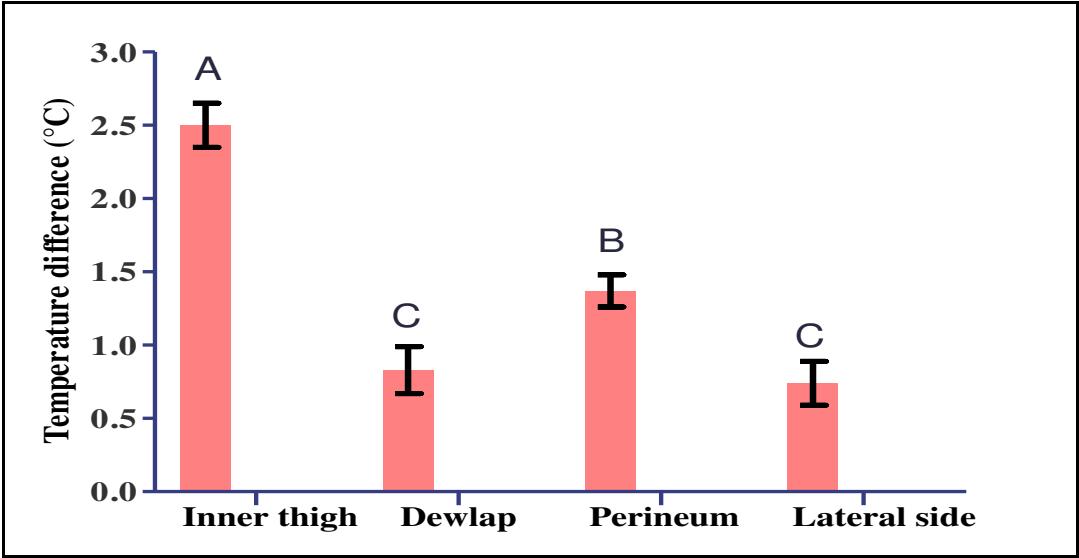


Fig 3: Difference between the tick and skin surface temperature (°C) in various region of animal body; Bars with different superscripts differs significantly ^{AB}(*p*<0.01)

Fore noon	IRT Palette 1	IRT Palette 4	IRT Palette 11	Image-J
6 AM				
7 AM				
8 AM				
9 AM				
10 AM				
11 AM				
12 Noon				

Fig 4: IRT Palette 1, Palette 4, Palette 11 and Image-J analysis photographs of inner thigh region

Conclusion

The present study has identified 6:00 to 10:00 A.M. as the optimal time for capturing infrared thermography (IRT) images for tick identification in crossbred cattle. During this period, the thermal contrast between ticks and the host skin was highest, facilitating accurate detection and quantification. As the ambient temperature increased toward midday, reduced thermal differentiation adversely affected tick identification. Therefore, early-morning IRT imaging, combined with automated image analysis, offers a reliable, non-invasive approach for standardized tick assessment under tropical conditions.

Conflict of Interest

The authors have no conflicts of interest.

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