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Physiological Responses of Bali Cattle Associated with Temperature-Humidity Index under Tropical Environmental Conditions

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Abstract
The temperature-humidity index (THI) is an indicator that combines air temperature and humidity to assess the level of comfort or heat stress in livestock. This study aims to evaluate the relationship between the physiological response of Bali cattle to THI in tropical areas. A total of 20 Bali cattle (10 males and 10 females) were observed in the morning and afternoon. Microclimatic data (temperature, humidity, THI) and physiological responses, including rectal temperature (RT), skin surface temperature (ST), respiratory rate (RR), and heart rate (HR), were analysed using the General Linear Model (GLM) with repeated measures and linear regression. The results showed significant differences ($P<0.01$) between morning and afternoon. THI increased from 77.34 ± 0.82 (mild heat stress) in the morning to 86.04 ± 1.20 (severe heat stress) in the afternoon. Physiological responses increased significantly in the afternoon, with males showing higher RT and RR than females. Regression analysis indicated that THI was closely related to RT ($R^2 = 0.8822$), RR ($R^2 = 0.8065$), and HR ($R^2 = 0.9454$), while ST showed a moderate relationship ($R^2 = 0.5503$). It can be concluded that Bali cattle are sensitive to tropical climate fluctuations, and THI can be used as an indicator to assess heat stress levels in Bali cattle.

Keyword: *Bali cattle; heat stress; physiological response; THI; tropical environment*

1. Introduction

Heat stress is one of the problems faced by livestock in tropical regions because it can affect their welfare, health, and productivity, as well as cause significant financial losses [1;2]. Several physiological characteristics, such as rectal temperature (RT), skin surface temperature (ST), respiratory rate (RR), and heart rate (HR), are often used to assess heat stress in cattle [3]. Understanding how tropical livestock thermoregulation works is crucial in designing effective mitigation strategies to minimize the impact of heat stress and ensure sustainable livestock production [4;5].

Bali cattle are one of the most popular beef cattle breeds cultivated in several regions of Indonesia (Saili, 2020), contributing substantially to national food security [6]. Their adaptability to tropical climates, ability to utilize low-quality feed, and relatively high physical endurance compared to other local cattle are some of the main advantages of Bali cattle [7]. Farmers widely raise Bali cattle in areas like Kutai Kartanegara Regency, East Kalimantan. These conditions can cause heat stress in livestock by disrupting the balance between heat production and heat dissipation, which is reflected in physiological conditions such as increased body temperature (RT), skin temperature (ST), respiratory rate (RR), and heart rate (HR).

In tropical livestock studies, diurnal variations in ambient conditions are significant factors to consider. In the morning, the ambient temperature (Ta) is relatively lower, and the relative humidity (RH) is higher, which results in more comfortable conditions for animals. In contrast, in the afternoon, there is a significant increase in temperature due to solar radiation, accompanied by a decrease in humidity [8]. This combination causes an increase in the Temperature-Humidity Index (THI), which reflects the level of heat stress in livestock. The relationship between environmental conditions and physiological responses plays an important role in the productivity and welfare of animals in tropical regions, especially local livestock [9], such as Bali cattle. Understanding physiological responses to THI is crucial in formulating sustainable management strategies in areas with high temperatures and humidity [10].

Therefore, this study aims to analyze the physiological responses of Bali cattle to THI under tropical environmental conditions. These findings provide perspectives on the adaptation mechanisms of Bali cattle and the implications of heat stress for livestock management in tropical conditions.

2. Materials and Methods

The study was conducted at a cattle farm located in Manunggal Jaya Hamlet, Tani Bhakti Village, Loa Janan Subdistrict, Kutai Kartanegara Regency, East Kalimantan, Indonesia (0°39'49.3"S 117°06'21.1"E, 23 masl), which has a humid tropical climate. A 2 × 2 factorial design with repeated measures was implemented, consisting of two factors: Factor A was the sex of Bali cattle (male and female), and Factor B was the time of measurement (morning and afternoon).

The experimental animals used were 20 Bali cattle aged 4–5 years, grouped by sex, consisting of 10 males with an average body weight of 300 kg and 10 females with an average body weight of 250 kg. The feed provided was a combination of forage and concentrate at a ratio of 70:30 (dry matter basis). Forage consisted of fresh Pennisetum purpureum and Leucaena leucocephala leaves at a ratio of 80:20, while the concentrate consisted of 60% rice bran, 25% ground corn, 10% coconut meal, 3% molasses, and 2% mineral-vitamin premix. The target feed intake was set at 2.5% of body weight per head per day, so that male cattle received approximately 26.25 kg of fresh forage and 2.56 kg of concentrate, while female cattle received approximately 21.88 kg of fresh forage and 2.13 kg of concentrate. Feed was given twice daily, and drinking water was provided ad libitum. Each animal was measured both in the morning and in the afternoon, providing two measurement points per head.

An intensive rearing system was applied, in which cattle were kept fully in confinement and not allowed to graze. The housing used was an open-house type with a gable roof made of zinc material. Each animal was housed in an individual pen measuring 1 × 1.5 m, arranged in a head-to-head layout with a central alley to facilitate maintenance activities. The floor of each pen was covered with rubber mats to provide comfort and prevent injury, and each pen was equipped with a feeding and drinking trough.

The parameters observed included microclimate conditions and physiological responses. Ambient temperature (Ta) and relative humidity (RH) were measured simultaneously with physiological observations in the morning and afternoon using a thermohygrometer (Thermohygrometer Clock HTC-2) placed at the measurement site. The Temperature–Humidity Index (THI), as an indicator of environmental comfort level for livestock, was calculated using the formula adapted from the National Resource Center (NRC) [8]:

$$THI = (1.8 \times Ta + 32) - (0.55 - 0.0055 \times RH) \times (1.8 \times Ta - 26)$$

where Ta = ambient temperature (°C) and RH = relative humidity (%).

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Physiological parameters were measured twice, in the morning (06:00–08:00) and in the afternoon (12:00–14:00), following the procedure previously conducted by [8]. Rectal temperature (RT) was measured using an Omron digital thermometer model MC-341 (Omron Healthcare Co. Ltd, Japan) inserted into the rectum to a depth of 6–7 cm until a "beep" sound was heard, with values expressed in °C. Skin surface temperature (ST) was measured non-invasively using a Sinocare infrared thermometer model AET-R1D1 (Sinocare, Indonesia) with ±0.2°C accuracy. Measurements were taken approximately ±0.5 cm from the skin surface at four points: chest (A), back (B), upper leg (C), and lower leg (D). ST was calculated using the formula from [11]:

$$ST=0.25(A+B)+0.32C+0.18D$$

With values expressed in °C. Respiration rate was measured by counting flank movements for one minute without disturbing the animals, expressed as breaths per minute (bpm).

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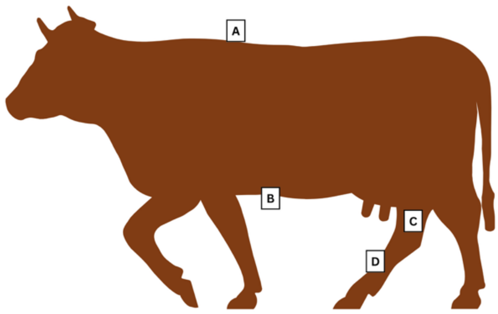

Figure 1. Skin Surface Temperature Measurement Point

Data analysis was carried out in several stages. The independent samples t-test was used to compare microclimate conditions between morning and afternoon. The effects of sex, time, and their interaction on physiological parameters were analyzed using the General Linear Model (GLM) with repeated measures. When a significant interaction was found, post hoc comparisons were conducted using Duncan's Multiple Range Test (DMRT). Statistical analyses were performed using IBM SPSS Statistics version 29. In addition, linear regression analysis was applied to determine the relationship between the Temperature–Humidity Index (THI) and physiological parameters, and the results were visualized using scatterplots with regression lines in Microsoft Excel.

All experimental procedures complied with animal welfare regulations and were approved by the Animal Ethics Committee of the Department of Animal Husbandry, Faculty of Agriculture, Mulawarman University.

3. Results and Discussion

The tropical climate is characterized by fluctuating temperatures and humidity throughout the day, which greatly affects the comfort and productivity of livestock [12], particularly Bali cattle. To assess thermal comfort levels, one commonly used measure is THI, which is a combination of Ta and RH. The higher the THI value, the greater the heat stress experienced by livestock, which can affect their physiological response and production performance. The results of microclimate measurements are presented in Table 1.

Table 1. Microclimate Conditions at the Research Location

Microclimate	Morning	Afternoon
T_a (°C)	25.75 ± 0.53^a	33.22 ± 0.42^b
RH (%)	91.05 ± 0.98^a	69.05 ± 5.09^b
THI	77.34 ± 0.82^a	86.04 ± 1.20^b
Livestock Comfort Category	Mild Stress	Severe Stress

Description: Different superscripts (a,b) within the same row indicate significant differences (P < 0.01).

Measurements of microclimate conditions inside the house showed highly significant differences (P<0.01) between morning and afternoon for Ta, RH, and THI measurements. Ambient temperature (Ta) was much lower in the morning than in the afternoon, when it increased significantly. In contrast, RH was higher in the morning but decreased markedly in the afternoon. The THI also shows a clear difference, with a higher value during the afternoon compared to the morning. In this study, the Ta was recorded at 25.75 ± 0.53°C in the morning and increased to 33.22 ± 0.42°C in the afternoon, with RH at 91.05 ± 0.98% in the morning and decreasing sharply to 69.05 ± 5.09% in the afternoon. Based on these conditions, the THI value in the morning was 77.34 ± 0.82, and in the afternoon it was 86.04 ± 1.20, indicating mild heat stress and severe heat stress, respectively [13].

An increase in Ta and a decrease in RH occur when solar radiation intensity is at its peak [8]. High solar radiation intensity increases the heat load around the house area, causing Ta to increase. At the same time, solar radiation also accelerates evaporation from the ground, walls, and floor of the house, which decreases RH. These conditions will exacerbate heat stress in livestock. Microclimate data, such as Ta and RH, are used to calculate the THI value, which aims to examine the impact of heat stress on livestock [14; 8;15]. Heat stress is a condition in which livestock are unable to maintain thermal homeostasis due to external factors such as unsupportive Ta and RH [16]. This condition also harms productivity because livestock use energy that should be used for production purposes, but is diverted for adaptation to environmental changes [17].

Physiological Response of Bali Cattle

The physiological response of Bali cattle in this study was observed through RT, ST, RR, and HR. The physiological responses of Bali cattle are presented in Table 2. The results indicated that the time of observation (morning vs. afternoon) had a dominant effect on all physiological parameters (P<0.01), with RT, ST, RR, and HR increasing during the afternoon in line with higher THI conditions (severe heat stress category). RT, ST, RR, and HR in the morning were at 36.69±0.50°C, 35.82±0.62°C, 22.95±0.83 bpm, and 54.74±1.29 bpm, respectively. Then they increased sharply during the day to 38.74±0.10°C, 36.75±0.18°C, 27.75±1.37 bpm, and 79.20±1.74 bpm, respectively. This finding aligns with [18], who discovered that an elevation in Ta results in increased RT, RR, and HR as adaptive responses to heat stress.

As THI increases, body temperature (RT) also tends to increase. This trend occurs because animals cannot release internal body heat, causing it to accumulate. An increase in RT is a key indicator of heat stress in Bali cattle, as it indicates that they are unable to dissipate heat effectively when THI is high [19]. At the same time, ST increases because peripheral vasodilation increases blood flow to the skin tissue, which helps the body transfer heat to the environment. This process does not function optimally when THI is too high because the temperature difference between the body and the environment becomes smaller [20]. [21] explain that an increase in ST due to heat stress can interfere with the ability of cattle to regulate their body's thermal balance.

When THI increases, RR responds by increasing to strengthen the cooling mechanism through evaporation via the respiratory tract [22]. Furthermore, an increase in THI causes an increase in HR, which is the result of increased circulatory activity that transfers heat to the body surface. [23] mention that an increase in HR can be a compensation for vasodilation and increased peripheral blood flow during heat stress. Overall, physiological parameters such as RT, ST, RR, and HR are interrelated in the body's thermoregulatory mechanism, which is to maintain a balance between metabolic heat production and heat release through radiation, convection, conduction, and evaporation [18; 20].

Table 2. Physiological Response of Bali Cattle

Parameters	Factor	Group	Average ± SD	Significance
RT (°C)	Sex	Male	37.89±1.02	**
		Female	37.54±1.17	
	Time	Pagi	36.69±0.50	**
		Siang	38.74±0.10	
	Sex*Time	Male – Morning	36.96±0.54	ns
		Male – Afternoon	38.82±0.08	
		Female – Morning	36.42±0.30	
		Female – Afternoon	38.66±0.05	
ST (°C)	Sex	Male	36.30± 0.61	ns
		Female	36.27±0.71	
	Time	Pagi	35.82±0.62	**
		Siang	36.75±0.18	
	Sex*Time	Male – Morning	35.90±0.65	ns
		Male – Afternoon	36.69±0.10	
		Female – Morning	35.73± 0.61	
		Female – Afternoon	36.81±0.23	
RR (napas/menit)	Sex	Male	26.05±2.94	**
		Female	24.65±2.21	
	Time	Pagi	22.95±0.83	**
		Siang	27.75±1.37	
	Sex*Time	Male – Morning	23.3±0.82[a]	*
		Male – Afternoon	28.8±1.03[c]	
		Female – Morning	22.6±0.70[a]	
		Female – Afternoon	26.7±0.67[b]	
HR (denyut/menit)	Sex	Male	68.05±12.83	ns
		Female	65.90±12.34	
	Time	Pagi	54.74±1.29	**
		Siang	79.20±1.74	
	Sex*Time	Male – Morning	55.6±1.17	ns
		Male – Afternoon	80.5±1.27	
		Female – Morning	53.9±0.74	
		Female – Afternoon	77.90± 0.99	

Description: ns = not significant (P>0.05), * = Significant (P<0.05), ** = very significant (P<0.01)
Different superscripts (a,b) within the same column indicate significant differences (P < 0.01).

Sex differences also had a significant effect on RT and RR (P<0.01) but not on ST and HR (P>0.05). In this study, RT in males was higher at 37.89 ± 1.02°C, while in females it was 37.54 ± 1.17°C. In addition, RR in males was 26.05 ± 2.94 bpm, and in females it was 24.65 ± 2.21 bpm. A previous study on Madura cattle reported similar findings, with males showing greater changes in RT and RR than females [24]. This phenomenon can be associated with variation in basal metabolism and hormonal activity between sexes [25; 26]. [27] explain that anthropometric differences (body size, surface area to mass ratio) between sexes influence heat dissipation efficiency, where females have a higher surface area to mass ratio may be effective for heat dissipation. This is consistent with the biophysical/anthropometric model by [28], which shows a greater risk of overheating in male mammals due to the size/mass trade-off against surface area ratio, supporting the interpretation that males are more susceptible to metabolic heat accumulation.

The absence of differences in ST and HR (P>0.05) likely results from cardiovascular regulation and skin surface responses being more influenced by environmental than internal bodily factors. Since both sexes were under the same environmental conditions, ST tended to be similar despite differences in RT. The skin is more susceptible to fluctuations due to external factors, such as wind, solar radiation, and humidity, so that the influence of sex is not very noticeable [30;31]. The skin itself functions as a heat radiator that allows heat to be released into the environment through radiation, convection, and evaporation [20]. This mechanism occurs when blood vessels dilate (vasodilation) and sweat. Vasodilation occurs when the heart pumps more blood to the capillaries on the skin's surface to transfer heat to the external environment [20;31]. If this mechanism is not effective enough to cool the body, for example, when the external temperature is higher than the body temperature, the brain will instruct the sweat glands to release fluid to the skin's surface. The sweat then evaporates, which helps the body shed excess heat by changing from liquid to vapor [32]. As a result, both males and females exhibit similar physiological mechanisms in response to heat stress, and HR does not show significant differences between the two sexes.

The interaction between sex and time showed a significant effect ($P<0.05$) on RR parameters. The study showed that males were more sensitive to increases in THI than females, resulting in a higher respiratory response during the daytime. Males have a higher RR than females, with a sharper increase from morning to afternoon (23.3 to 28.8 bpm), while females increase more moderately (22.6 to 26.7 bpm). This scenario proves that male cattle are more sensitive to THI fluctuations and rely more on evaporation through respiration to maintain physiological balance. This difference can be attributed to the fact that male cattle have larger bodies and more muscle, which causes them to generate more metabolic heat [27].

The Relationship Between THI and Livestock Physiology

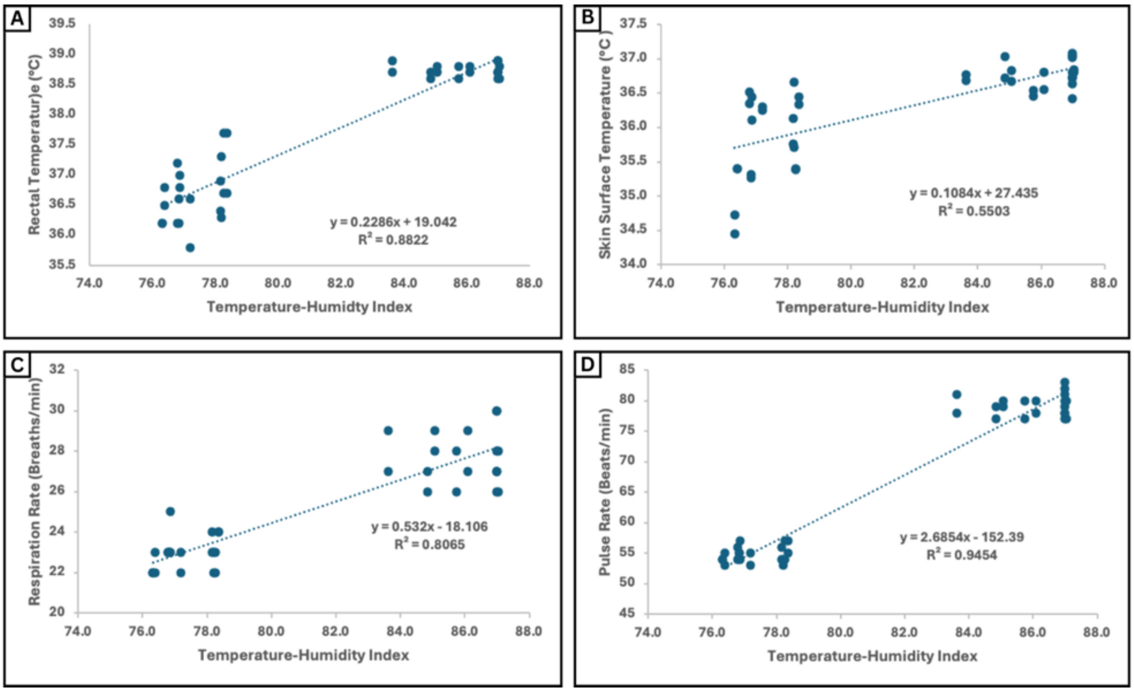

Figure 2. Scatterplot of THI vs. Physiological Parameters: a) THI with rectal temperature; b) THI with skin surface temperature; c) THI with respiratory rate; and d) THI with heart rate

The results of the linear regression analysis between the Temperature-Humidity Index (THI) and physiological parameters show variations in the degree of correlation (Figure 2). The regression equation obtained describes the direction and magnitude of change in each physiological parameter in response to an increase in THI, while the coefficient of determination

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(R^2) value indicates the extent to which variations in physiological parameters can be explained by THI.

Changes in THI significantly affect RT (0.8822), RR (0.8065), and especially HR (0.9454) due to their high R^2 values. In other words, the environmental conditions reflected in the THI may explain more than 80% of the physiological variation in that parameter. The R^2 value is higher than the study by [33] on Korean local calves, which found R^2 values of 0.8368 for THI against HR and 0.6162 for RT. This study indicated that cattle exposed to high THI had higher RT and HR than cattle exposed to low THI [34]. [35] Also found a similar pattern in beef cattle in Indonesia, where higher THI during the day was followed by an increase in RT and HR.

A study in Germany on dairy cows also found that HR and RR showed a significant increase after THI exceeded a certain threshold, although the response was lower than our findings [36]. The results may indicate that the local cows studied had different thermoregulatory adaptations or that the THI range observed in this study included more extreme conditions, resulting in a greater response. This is in line with the concept that increases in environmental temperature and humidity increase the body's heat load, which is then responded to through increases in internal temperature, respiratory rate, and cardiovascular system activity [37].

On the other hand, ST has a lower R^2 value (0.5503), which means that there are other factors influencing it besides THI. [38] emphasized that skin thickness, coat color, and evaporation mechanisms also influence livestock response to heat stress. It is known that factors such as peripheral blood flow, the size and activity of sweat glands, and coat thickness and color can influence how heat is released. [39] explain in their study of Brahman cattle that having physiological advantages, including increased blood flow from the centre of the body to the skin surface, larger sweat glands, and shorter, lighter-coloured hair, contributes to reduced heat radiation absorption.

In addition to providing a relationship, linear regression also allows us to estimate how much physiological parameters will increase each time THI increases by one unit. The equation shows that every 1 THI increase will increase RT by 0.23°C ($y = 0.2286x + 19.042$), ST by 0.21°C ($y = 0.2084x + 27.435$), RR by 0.53 breaths/minute ($y = 0.532x – 18.106$), and HR by 2.69 bpm ($y = 2.6854x – 152.39$). These results indicate that HR (cardiovascular system) is most sensitive to changes in THI, followed by RR and RT. ST is less responsive, which may be due to factors other than THI, such as peripheral blood flow, skin thickness, hair colour, and evaporation mechanisms.

4. Conclusion

The results indicated that microclimate conditions, particularly an increase in THI during the day, caused severe heat stress (THI = 86.04 ± 1.20) in Bali cattle. Physiological responses included increases in RT, ST, RR, and HR, which were higher during the day, with significant differences between males and females in RT and RR parameters. Regression analysis shows that THI is closely related to RT ($R^2 = 0.8822$), RR ($R^2 = 0.8065$), and HR ($R^2 = 0.9454$), while ST is lower ($R^2 = 0.5503$), indicating that there are other factors influencing THI. These findings confirm that Bali cattle are sensitive to tropical climate fluctuations and that THI can be used as an important indicator in assessing heat stress.

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