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Elucidating the Complex Relationship between the Neuroendocrine System and Environmental Stresses Identify by Possible Markers of Heat Stress in Cross Bred Calves

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ABSTRACT

Background: The behavioral, physiological and hormonal reactions of crossbred calves to various shade materials are the primary foci of this investigation into the multi-faceted effects of heat stress. The goal of research is to identify possible markers of heat stress resistance by elucidating the complex relationship between the neuroendocrine system and environmental stresses.

Methods: By making various measurements regarding the calves' physiological responses to heat stress and the functions of various kinds of shade materials.

Result: Behavioral observations showed that various shade materials alleviated stress in different ways. In the open paddock, the crossbred calves were able to maintain normal physiological, biochemical and behavioral responses because of the favorable microenvironment created by the availability of agro-net and thatch as shade material throughout the summer. The effects of shade on metabolic parameters were shown by biochemical studies, which shed light on the physiological adaptations of the calves. Moreover, evaluations of hormones revealed complex hormonal modulations as adaptive processes. This study concluded that agro-net and other novel shade materials might be considered for use as summer cow shade; however, the material's longevity has to be studied.

Key words: Behavioural, Biochemical, Cross Bred Calves, Heat stress, Hormonal Reaction, Shade Material.

INTRODUCTION

The agricultural sector faces with new and exceptional challenges in maintaining cattle against the effects of increasing global temperatures and climate change. Crossbred calves are more susceptible to the damaging effects of heat stress on their bodies and minds due to their genetic makeup. Understanding the complex ways that crossbred calves react to heat stress is crucial, especially as the cattle industry deals with the effects of climate change (Johnson *et al.,* 2018). The complex relationship between the hormonal, physiological and behavioural reactions in crossbred calves kept in hot environments, with the goal of improving management techniques and the animals' well-being as a whole (Brown *et al.,* 2021). Crossbred calves are among the most sensitive animal groups due to the risk for greater physiological strain caused from high temperatures (Banerjee and Ashutosh, 2011). The entire performance and production of these animals are compromised and their wellbeing is significantly threatened, by heat stress. A number of management solutions have been developed to address this pressing issue; one such strategy is the use of shade materials, which have shown promise in reducing the negative impacts of heat stress (Jat and Yadav, 2010).

Heatwaves are becoming more frequent and severe as a result of global warming, which is a major concern for the well-being of animals. Crossbred calves, an essential aspect of the livestock sector, are more susceptible to heat stress, hence it is essential to thoroughly examine their adaption mechanisms and responses. One of the most basic ways to protect cattle from heat stress is to provide them shade. The efficacy of various shade materials in reducing heat stress is still up for debate, however the purpose of

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this research is to examine the effects of different types of shade on the physiological, hormonal and behavioral components of heat stress in crossbred calves (Roy and Chatterjee, 2010). It is common for crossbred calves to have altered sleep and activity cycles as a result of heat stress. The health and happiness of crossbred calves is greatly influenced by their social relationships. The social stresses that make rising temperatures worse may be better understood by delving into these processes (Smith *et al.,* 2016). Calves of different breeds have changes in energy expenditure and food consumption as a result of the metabolic load that heat stress causes. The cellular integrity of crossbred calves may be put at risk by elevated temperatures, which can promote oxidative stress. To better understand these animals' resistance to oxidative damage and possible intervention strategies, it is crucial to investigate their antioxidant defence systems.

The stress hormone cortisol is a good measure of how animals' bodies react to stress. One way to quantify the stress levels experienced by crossbred calves exposed to heat stress is to investigate cortisol dynamics; this will also help in measuring the effectiveness of shade materials in mitigating stress (Walker *et al.,* 2019). When it comes to controlling body temperature, thyroid hormones are key regulators of both metabolic rate and heat dissipation pathways. By analyzing how thyroid hormone levels change in response to heat stress, we may learn more about the processes that regulate body temperature.

One common and effective way to help animals deal with heat stress is to provide them shade. Shaded areas for cattle have been constructed using a variety of materials, including native flora, man-made shelters and novel fabric-based constructions. The optimal management of shade and the safety of crossbred calves in hot climates depend on our ability to understand how various shade materials affect the biochemical, hormonal and behavioural reactions of these animals (Martinez *et al.,* 2018).

This research explores the wide variety of shade materials that may be used to control cattle, including both man-made buildings and natural plants, as well as materials that have been specifically engineered to dissipate heat. Different materials have different properties that may affect how crossbred calves react hormonally, biochemically and behaviourally. Investigating the complex relationship between various shading components and the animals' physiological reactions (Foster *et al.,* 2019).

This study looks at how various types of shade affect the physiological, hormonal and behavioral reactions of crossbred calves while they are hot. Heat stress is a major problem for cattle since it reduces their health and production. In order to create efficient management techniques, it is essential to understand the effects of shade materials on physiological indicators. The adaptive techniques that the calves use to cope with stress may be better understood via behavioral observations. To further understand how cells react to heat stress, biochemical studies will evaluate important markers like metabolic profiles and oxidative stress indicators. Researchers and ranchers can improve animal welfare and production by finding shade materials that make animals more comfortable and lessen the impact of biochemical and hormonal abnormalities. This study lays the groundwork for the creation of realistic and affordable strategies to reduce heat stress in crossbred calves, which has important ramifications for sustainable agriculture and animal husbandry.

Particularly in crossbred calves, heat stress is a major risk to cross bred calves health and production. Although shade is an important tactic for reducing heat stress, little is understood on how various materials affect the hormonal, behavioral and physiological reactions in crossbred calves. To fill this knowledge vacuum, this study will examine how different types of shade affect the physiological markers, hormonal profiles and behavioral patterns of crossbred calves. Finding the best materials for shade can improve the animals' health and welfare and help with sustainable livestock management. Implications for the livestock sector are significant since the research sheds light on how to best protect crossbred calves from heat stress and how to increase their resilience in a variety of environments.

MATERIALS AND METHODS

The investigation included 28 Vrindavani crossbred calves (without biasness in sex) were chosen at random beginning three days after birth. The cows after parturition, calves fed with formulated feed instead of nursing them. The seven calves in each enclosure were randomly assigned to one of three groups according to their average birth weight. There was a combination of open and covered areas for each group's housing. What follows is a description of the methods utilized to shade covered areas under each treatment, including the specific materials employed.

T1: (Thatch shading roof).

T2: (Agro-net shading roof).

T3: (Asbestos with canvas shading roof).

T4: (Well-grown tree).

The east-west axis was the limiting factor in its length. For each treatment group, covering zones of 1.5 × 1 m were created using various shade materials. A welded wire mesh open paddock measuring 2 by 1 meter was constructed. Brick flooring and 2.5-meter-high eaves characterize this shelter. During the first two days of their existence, the mother took care of the new born calves. After three days, the calves were split in halves and kept as per the protocol. The weight of each calf determined how much whole milk to give them. One tenth of body weight between days 4 and 28, one fifteenth between days 29 and 42 and one twenty-twentieth between days 43 and 56. The amount of milk needed was provided using the pail technique and any excess was also noted.

The first week and thereafter saw the introduction of green fodder and calf starter. The most widely grown green crops in India, including Berseem, maize and jowar, were among the most frequent green fodder crops offered. Feed Technology Unit-made calf starter-a mixture of crushed maize, soya bean oil cake, wheat bran, mineral combination and common salt-was used in the experiment. All of the calves were grown in sanitary conditions with plentyof supervision during the investigation. Deworming should be done when the calves were about two weeks old. After that, disbudding was accompanied with precautions in the next week. Throughout the duration of the experiment, we provide, sufficient food, water containers and the area around the animals very clean and also sure to maintain the enclosures. Every day, the pens were disinfected and every measure was taken to avoid the spread of infectious illnesses. During the duration of the trial, the calves were bathed every morning.

Every day at 9:00 and 1400 hrs, researchers recorded both the macro- and microclimatic parameters. The former included the weather in the study location, while the latter referred to the environment within the calves' enclosure. We counted the day's hottest and lowest temperatures using special thermometers. The daily relative humidity was recorded using a psychrometric chart. Suspended on a thread in the enclosed space below the ceiling, it read dry and wet bulbs. In order to calculate the temperature humidity index (THI):

THI= 0.72 (wet bulb temperature + dry bulb temperature) + 40.6

Upon initiation of the study, 3 ml of blood samples were collected from jugular vein of each calf at 0, 15, 30, 45, 60, 75 and 90 days. Using Drabkin's method, the blood hemoglobin level was found. An assay provided by Erba Diagnostics Mannheim GmbH, Germany, was used to estimate serum biochemical and cortisol hormone levels.

For three consecutive days, once a month, all animals in each group had their continuous activities recorded using closed circuit television from 1000 to 1700 hours after milk feeding. The study relied on specialized coding sheets to record and classify the duration of each behavioral activity. The research's treatment and control groups were compared using analysis of variance (ANOVA) in SPSS 22. The data was aggregated from the various sources. The data were examined at 14-day intervals after being compressed throughout the process.

RESULTS AND DISCUSSION

Behavioural response analysis

The daily activities of crossbred calves are shown in Table 1 for each summer season. Over a 7-hour period (1000-1700 h), the calves' exposure to shade and open areas, as well as the total time spent in various behavioral activities, are taken into account, especially the overall impact of shade material.

There was a significant difference in the amount of time that the calves in the T2 group spent eating, ruminating, resting, sleeping and playing compared to the other groups ($P<0.05$). Similarly, the T2 group was on their feet and moving about for much less time. The times it took for defecation, T2 and T3 were 1.20±0.07, 1.40 ±0.15, 1.00± 0.06 and 1.44±0.08 minutes, respectively. In contrast, urination for T4, T3 and T4 took 1.13±0.07, 1.11±0.08, 0.99 ±0.06 and 1.56±0.07 minutes, respectively. With the exception of T3, all of the other groups' calves participated in each activity more in the shaded area than in the open area.

Biochemical response analysis

Haematological response

Table 2 displays the hemoglobin levels of crossbred calves exposed to various shading materials. Calves typically have normal hemoglobin levels between 5.6 and 12.5 g/dl. During the fourth 14-day period of the trial, T2 had considerably higher haemoglobin levels than T3 ($P<.05$).

Biochemical response

As shown in Table 3 and 4, different biochemical parameters were affected by different types of shade materials. Blood glucose values were higher in T2 compared to the other calf groups ($P<.05$). The ALP was noticeably greater in T4 ($P<.05$) compared to T2, T3 and T2. In addition, The total ALP value ranged from 95 to 196 IU/L, which is within the usual range of 64 to 222 IU/L. All of the animalss have SGOT readings lower than the typical range of 26–58 IU/L. Nevertheless, in comparison to T2 and T1, the SGOT level in T4 and T3 was noticeably greater ($P<.05$), correspondingly.

Hormonal response analysis

The results of a 14-day research that looked at how different types of shade affected cortisol and thyroxin (T3 and T4) are shown in Table 5. The blood cortisol level was considerably greater at the start and end of the 14-day period for all groups. The total cortisol level ranged from 7 to 10 nmol/L, which is greater than the predicted range of "4-6.5 nmol/L".

Temperatures in T3 and T4 reached their maximums. It is possible that the shade material utilized in T3 couldn't reduce the radiative heat load; yet, it did absorb the heat and transmit it to the shed's microenvironment, so everything is in accord. In contrast to other shade structures, shade tree (T4) may have offered less shelter from direct sunlight (Rodriguez *et al.,* 2016).In terms of protective potential, the agro-net shading material outperformed all others based on T2 because it maintained the mean maximum temperature beneath the shadow structure. The current results corroborate earlier studies that found that thatch and mud plaster roofs, as compared to loose dwellings coated with an asbestos sheet, had generally cooler temperatures ($P<.05$). Based on research, a home with a tile roof had higher maximum temperatures and low minimum temperatures compared to with an asbestos roof (Taylor *et al.,* 2020).

One possible explanation for the lower hemoglobin levels in T3 compared to T2 is haemodilution, which involves increasing the circulation of water to cool it down through evaporation (Banerjee and Ashutosh, 2011). It has been proposed that an exceptionally high hemoglobin value could be a sign of exceptional heat tolerance, as this trait is linked to a high degree of adaptation to extremely hot and cold environments.Calves in the T4 and T3 groups exhibited greater activity of serum SGOT and SGPT compared to the other groups. Heat stress weakens the physiological and biochemical homeostasis processes, which is why this happens under support of the present findings, Brijesh (2012) discovered that serum SGOT and SGPT activity increased under various heat stress conditions, which is consistent with Baker *et al.* (2017).

Glycogenolysis and gluconeogenesis were both inhibited in calves during hot weather (Bahga *et al.,* 2009). The present results are in agreement with Baker *et al*. (2017), that buffalo calves given sprinklers and fans to alleviate the summer heat stress had significantly elevated blood glucose levels. Consistent with the current results, Baker *et al.* (2017) also found that children kept outside all summer had substantially higher cortisol levels, but they did not find a statistically significant difference in levels between the three treatment groups (thatch, agro-net and tree). While it was shown that T3 and T4 harmone concentrations decreased as heat exposure increased, this trend did not achieve statistical significance (Kamal *et al.,* 2014).

Heat stress may promote alkalosis *via* increased alveolar ventilation, which in turn raises ALP activity at T3 and T4. The increased activity seen in this study might be attributed to its function in controlling animal energy production, homeostasis and cell division and development. According to what was predicted in (Bahga, 2007), this result is consistent.

It is possible that the calves' increased eating time was caused by the pleasant microclimatic conditions created by the shade material in T2. Nevertheless, it is thought that a notable variation in serum biochemical level is caused, in part, by calves in T3 attempting to deal with heat load. Sanker *et al.* (2012) found that animals whose

Table 1: The average±SE of Rumination, resting, standing, moving, sleeping, grooming, play licking, cross licking of the calves' and total behavioral activity in minutes.

Behaviours		(T1)	(T2)	(T3)	(T4)
Rumination	Shade	77.01±1.71b	87.87±0.86c	32.13±0.78a	73.93±0.89b
	Open	22.15±0.63a	20.41±0.60a	62.24±1.52b	20.42±0.67a
	Total	89.17±1.89B	78.29±0.82C	64.36±1.65A	84.33±0.75A
Resting	Shade	77.55±1.56c	65.78±2.07c	34.16±0.70a	72.17±0.82b
	Open	25.06±0.46ab	22.28±0.63a	46.62±1.72c	27.87±0.64b
	Total	72.58±1.47A	78.05±1.87B	70.79±1.82A	70.05±0.99A
Standing	Shade	31.31±0.96b	25.46±0.57a	35.46±0.44c	30.78±0.85b
	Open	28.77±0.68c	7.64±0.6a	28.52±0.76c	23.42±0.48b
	Total	30.07±1.14c	32.09±0.67a	53.97±0.71c	44.19±0.86b
Moving	Shade	28.33±0.78	25.32±0.55	27.22±1.06	26.94±0.58
	Open	24.68±0.72b	20.97±0.58a	7.98±0.75a	31.73±0.75c
	Total	43.02±1.25b	36.26±0.90a	37.17±1.52a	47.67±0.82c
Sleeping	Shade	40.96±0.94c	47.36±1.38d	26.02±0.57a	36.81±0.38b
	Open	7.36±0.68b	5.11±0.36a	26.57±0.80c	9.45±0.44b
	Total	27.31±0.78B	31.43±1.47C	22.58±0.78A	45.24±0.75AB
Play	Shade	23.95±0.77a	15.34±1.33b	21.61±0.56a	22.66±0.51a
	Open	24.12±0.70c	9.32±0.31a	21.11±0.67b	22.61±0.55bc
	Total	38.06±1.11b	43.67±1.58c	22.71±0.94a	35.27±0.43ab
Grooming	Shade	5.58±0.37c	4.67±0.26b	5.82±0.26c	3.57±0.30a
	Open	3.56±0.16a	2.89±.22a	5.14±0.37b	3.24±0.08a
	Total	8.15±0.34b	4.55±0.31a	9.95±0.47c	5.81±0.26a
Licking	Shade	3.47±0.27b	1.47±0.21a	3.17±0.28b	1.07±0.14b
	Open	3.29±0.27b	2.78±0.09a	5.68±0.26c	1.37±.12b
	Total	5.77±0.23b	4.25±0.25a	7.86±0.41c	3.45±0.21b
Cross-sucking	Shade	2.57±0.35a	2.45±0.06a	8.42±0.08b	1.23±0.08b
	Open	3.17±0.34a	1.83±0.07a	9.74±0.23b	1.27±0.10a
	Total	4.77±0.31a	4.27±0.20a	10.18±0.25c	5.48±0.16b
Total time spent	Shade	347.73±1.75c	153.72±1.49d	135.94±2.03a	109.14±1.86b
	Open	78.14±1.85b	68.16±1.28a	166.57±2.73c	81.33±1.63b
Feeding	Shade	67.13±1.71ab	75.47±0.84c	65.94±1.44a	91.32±0.75bc
Drinking	Shade	20.28±0.73a	7.72±0.41a	23.66±0.53b	21.22±0.60a
Near water tank	Shade	24.43±0.64b	9.42±0.61a	35.91±0.62c	34.03±0.81c

Table 2: Calves' mean±SE of hemoglobin (g/dl) under various shade materials.

13-day period interval	(T1)	(T2)	(T3)	(T3)
Initially	9.17±0.96	9.23±0.67	7.69±0.32	9.21±1.11
I	9.91±0.93	9.09±0.93	9.63±0.61	9.09±0.31
II	9.97±0.76	9.99±0.36	9.79±0.37	9.97±0.69
III	11.62±0.77	11.33±0.79	9.79±0.39	11.31±0.66
IV	12.61±0.61ab	12.97±0.61b	11.02±0.60a	12.23±0.62ab
V	13.19±0.73	13.21±1.16	11.37±0.36	11.97±0.67
VI	13.36±0.72	13.37±1.13	11.93±0.32	13.21±0.36
Total	11.62±0.33	11.62±0.36	9.39±0.26	11.67±0.37

Table 3: Average± standard Error of blood biochemical assessment in calves with varying shade materials.

14-day interval	(T1)	(T2)	(T3)	(T4)
		Serum glucose (mg/dl)		
Initially	38.07±2.62	50.12±1.70	38.84±2.77	38.87±0.73
I	62.48±7.82	47.82±3.88	50.47±4.77	52.82±0.87
II	64.27±7.73	73.37±7.22	41.68±4.82	46.21±4.38
III	38.01±4.10a	72.87±7.11b	37.83±7.20a	32.86±2.77a
IV	37.26±5.33	51.13±3.82	37.32±6.11	31.27±2.63
V	42.17±7.78	47.68±4.87	41.28±4.01	37.67±2.28
VI	38.88±3.47	44.14±3.88	38.86±4.28	32.44±4.17
Total	40.32±1.72a	46.47±1.22b	38.48±1.63a	37.78±1.32a
		Total serum protein (g/dl)		
Initially	7.78±0.43	7.82±0.44	7.74±0.56	7.76±0.77
I	7.43±0.38	7.78±0.46	7.20±0.50	7.11±0.28
II	7.31±0.37	7.20±0.17	7.08±0.14	6.88±0.24
III	7.13±0.37	7.07±0.30	6.87±0.30	6.84±0.40
IV	6.88±0.32	6.87±0.33	6.27±0.27	6.76±0.38
V	6.62±0.38	6.73±0.37	6.11±0.28	6.77±0.27
VI	6.71±0.21	7.06±0.13	6.74±0.46	6.76±0.09
Total	7.12±0.16	7.18±0.11	6.86±0.15	6.87±0.13
		Serum albumin (g/dl)		
Initially	3.68±0.07	3.67±0.09	3.72±0.22	3.67±0.33
I	3.78±0.33	3.78±0.24	3.67±0.12	3.78±0.07
II	3.78±0.27	3.88±0.25	3.71±0.16	3.67±0.12
III	4.04±0.35	4.08±0.18	3.87±0.50	3.88±0.50
IV	3.62±0.24	3.82±0.35	3.78±0.29	3.44±0.29
V	3.77±0.27	3.88±0.28	3.73±0.17	3.68±0.27
VI	3.63±0.18	3.78±0.48	3.48±0.21	3.73±0.23
Total	3.76±0.12	3.86±0.16	3.68±0.13	3.68±0.06
		Serum globulin (g/dl)		
Initially	4.07±0.47	4.16±0.63	4.01±0.43	4.06±0.73
I	3.66±0.42	3.88±0.46	3.76±0.54	3.74±0.31
II	3.72±0.42	3.18±0.27	3.38±0.27	3.31±0.27
III	3.08±0.37	2.87±0.27	3.03±0.73	3.06±0.46
IV	3.18±0.27	3.16±0.27	2.67±0.36	3.13±0.40
V	2.87±0.36	2.74±0.38	2.37±0.34	2.87±0.32
VI	3.06±0.32	3.27±0.42	3.27±0.77	3.04±0.36
Total	3.34±0.12	3.33±0.20	3.18±0.17	3.27±0.17

Table 4: Average±SE of the serum enzyme levels in calves under various shadow structures.

14-day interval	(T1)	(T2)	(T3)	(T4)
		Alkaline phosphatase (IU/L)		
Initially	124.75±7.24	115.32±6.45	124.57±7.11	130.47±13.02
I	125.17±7.77	110.51±7.24	125.57±15.74	119.17±5.17
II	103.19±5.44	79.77±5.55	97.71±3.94	95.55±5.71
III	112.77±24.05	97.45±27.5	125.75±33.55	155.43±14.73
IV	110.47±21.42	75.55±15.3	117.73±17.92	144.57±21.09
V	152.71±15.41ab	132.17±14.9a	195.12±19.7b	193.77±11.0b
VI	147.05±9.17	129.04±10.7	171.57±24.90	177.74±25.25
Total	122.51±5.45ab	107.75±6.21a	157.35±21.9b	145.37±4.97b
		SGOT (IU/L)		
Initially	77.15±14.27	72.07±7.12	75.75±11.57	101.02±12.92
I	110.75±12.82ab	70.73±4.47a	127.55±13.0b	117.72±9.53b
II	73.35±20.50	79.57±7.54	115.15±14.75	117.45±14.27
III	137.55±11.23	74.52±7.73	135.37±37.00	125.27±7.13
IV	70.73±7.42	72.07±5.50	117.72±27.27	115.17±17.45
V	74.52±29.50a	79.57±13.00a	190.57±5.55b	174.39±30.4b
VI	97.25±14.32	97.51±12.71	103.55±11.75	133.77±17.25
Total	95.71±4.27a	75.34±2.77a	127.02±5.20b	127.28±4.72b
		SGPT (IU/L)		
Initially	34.35±5.35	40.72±5.77	40.57±5.25	41.73±4.83
I	39.14±4.83	35.77±7.55	37.91±7.44	37.77±7.50
II	33.75±7.00	34.11±5.50	40.93±5.12	37.15±5.55
III	39.92±5.57	35.37±3.79	45.73±2.77	49.49±4.99
IV	35.45±2.99	31.07±3.33	41.45±7.32	37.55±5.61
V	51.94±2.30	42.57±4.02	53.57±5.37	35.47±7.10
VI	33.32±2.39	32.57±2.35	47.52±3.87	44.57±8.52
Total	35.13±1.75	33.21±2.57	39.57±4.17	40.75±1.75

access to shade was greater (3.5 m^2 shade/animal) consumed more feed than those whose shade was less. T3 grouped calves drank more frequently during hot weather because their bodies takes less water through evaporative heat loss (sweat, panting, or respiration), which raised the osmolarity of the body's extracellular fluid. This, in turn, activated the hypothalamus's thirst center and prolonged the amount of time the calves drank.

During the heat, especially in areas without enough shade, the cattle consume more water than usual. During the third and fourth trimesters, calves spend more time in close proximity to a water tank likely because of the hot weather. Calves in the T3 and T4 groups may have taken measures, such as spending more time at the water trough, to alleviate heat stress because stress hormone more effective in summer season compare to spring season (Rai *et al.,* 2023). When the heat wave was at its worst, the proportion of unprotected beef cattle that congregated at the drinking well was twice as high as that of groups whose animals were provided with shade coverings of at least 3.5 square meters. The calf resting time was significantly shorter in T1, T3 and T4, which might be a sign of discomfort leading to decreased resting time and more standing time. The buffalo heifer's increased laying down time suggests that the sprinklers and fans are helping to alleviate some of the summer stress.

A 15% increase in heat load (THI=60-70) was associated with a 10% increase in standing time (13.8-15.3 h/day). Stookey and Watts (2007) proposed that cows stand for longer periods of time to enhance heat dissipation via increased skin exposure to wind and airflow. The pleasant microclimatic environment offered by the agro-net may explain why the calves in group T2 spend more time sleeping next to them. Furthermore, according to previous research with pastured cattle, the overall duration spent in shade was also comparable. In a similar vein, it was noted that cows spent a longer amount of time (P<.01) under shade that blocked a higher proportion of solar radiation throughout the day (50 and 99%), in comparison to 25% protection (Widowski, 2001).

CONCLUSION

This research examined the complex relationship between behavioral, biochemical and hormonal responses in crossbred calves that were exposed to different types of shade to prevent heat stress. Results highlight the importance of environmental factors in determining these animals' health. It is crucial to evaluate animal wellbeing

Table 5: Average± standard error of hormonal parameters in calves shaded by different materials.

13-day interval	(T1)	(T2)	(T3)	(T3)
		Cortisol (nM/L)		
Initially	13.62±0.69	13.63±0.96	13.02±1.31	13.39±0.91
I	11.95±2.39b	3.59±1.15a	9.35±2.80ab	5.96±1.62ab
Ⅱ	3.15±2.33	3.26±1.33	3.33±1.92	6.12±2.39
Ⅲ	3.99±1.39	3.66±1.19	3.61 ±1.92	9.39±2.40
IV	3.99±2.90	3.65±1.93	9.29±3.05	3.43±1.39
V	19.96±6.93	13.99±3.13	19.92±3.95	20.69±5.59
VI	15.69±3.92	10.02±2.26	19.22±3.35	11.13±2.50
Total	9.93±1.56	9.39±0.96	10.31±1.05	9.91±1.12
		T3 (nM/L)		
Initially	2.11±0.26	3.03±0.15	3.15±0.13	3.09±0.39
I	3.92±0.21	2.99±0.13	2.93±0.99	2.99±0.21
Ⅱ	3.55±0.26	2.62±0.25	2.61±0.20	2.31±0.32
Ⅲ	3.39±0.29	2.55±0.332	2.33±0.11	2.21±0.29
IV	2.62±0.39	2.89±0.52	2.56±0.29	2.33±0.13
V	2.35±0.30	2.59±0.39	2.19±0.35	2.19±0.19
VI	3.53±0.22	2.92±0.30	2.32±0.20	2.21±0.09
Total	2.62±0.11	2.99±0.19	2.59±0.26	2.35±0.20
		T3 (nM/L)		
Initially	59.89±3.51	50.12±2.59	54.93±3.00	55.31±9.83
I	57.33±6.89	63.55±3.66	54.99±9.30	54.99±9.96
Ⅱ	55.99±5.19	61.55±9.79	53.96±3.39	54.99±9.15
Ⅲ	53.99±5.99	60.39±9.59	54.55±3.32	53.03±13.92
IV	59.36±9.66	63.69±6.39	55.56±3.29	56.96±8.65
V	62.92±9.13	90.95±16.16	60.99±9.91	60.93±9.69
VI	60.19±5.66	69.12±9.69	59.93±9.29	59.96±9.96
Total	59.91±2.36	62.36±2.99	55.96±2.92	55.93±3.15

by taking behavioral markers into account in addition to physiological ones under heat stress conditions. Behavioral observations showed that various shade materials alleviated stress in different ways. In the open paddock, the crossbred calves were able to maintain normal physiological, biochemical and behavioral responses because of the favorable microenvironment created by the availability of agro-net and thatch as shade material throughout the summer. Moreover, evaluations of hormones revealed complex hormonal modulations as adaptive processes. Physiological values of calves housed beneath trees were similarly high, suggesting that this shade source was inadequate to provide a more favorable microenvironment for the young animals throughout the summer. It follows that agro-net and other novel shade materials might be considered for use as summer cow shade; however, the material's longevity has to be studied.

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Conflict of Interest

There is no conflict of interest among authors.

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