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ANIMAL PRODUCTION

Key points for the thermal comfort of water buffaloes in Eastern Amazon

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ABSTRACT: *This study presented relevant aspects about the Amazonian environment and how it impacts the thermal comfort of domestic buffaloes (Bubalus bubalis) raised in the Eastern Amazon. Furthermore, strategies for monitoring and mitigating animal heat stress are presented, based on research results with the species. Although domestic buffaloes are considered adaptable animals, exposure to intense solar radiation causes thermal discomfort. This condition is expressed in biophysical indicators, in metabolic, endocrine, behavioral responses, and in body thermographic patterns. Therefore, the biometeorological monitoring of production is crucial to support decision-making regarding environmental management strategies, genetic selection of thermotolerant individuals, and increase in animal welfare. Lastly, the use of silvopastoral systems can help to provide higher thermal comfort, which is a condition that directly impacts the productivity of milk and meat buffaloes when they are raised in tropical regions, such as in the Eastern Amazon.*
Key words: *ambience, Bubalus bubalis, thermal stress, thermoregulation, silvopastoral systems.*

Pontos-chave do conforto térmico de bubalinos na Amazônia Oriental

RESUMO: *Objetivou-se apresentar aspectos relevantes sobre o ambiente amazônico e como este impacta no conforto térmico de búfalos domésticos (Bubalus bubalis) criados na Amazônia Oriental. Adicionalmente, são apresentadas estratégias para monitoramento e mitigação do estresse térmico animal, a partir de resultados de pesquisa com a espécie. Apesar dos búfalos domésticos serem considerados animais adaptáveis, a exposição à intensa radiação solar provoca desconforto térmico. Essa condição é expressa em indicadores biofísicos, nas respostas metabólicas, endócrinas, comportamentais e nos padrões termográficos corporais. Assim, o monitoramento biometeorológico da produção é crucial para subsidiar tomadas de decisão em relação a estratégias de manejo ambiental, seleção genética de indivíduos termotolerantes e incremento do bem-estar animal. Por fim, o uso de sistemas silvipastoris pode auxiliar na oferta de maior conforto térmico, que é uma condição que impacta diretamente na produtividade de bubalinos de leite e carne, quando estes são criados em regiões tropicais, como na Amazônia Oriental.*
Palavras-chave: *ambiência, Bubalus bubalis, estresse térmico, termorregulação, sistemas silvipastoris.*

INTRODUCTION

Brazil has the largest herd of domestic buffaloes (*Bubalus bubalis*) in the Americas, with a population of 1.43 million animals and annual growth of 3.2% (IBGE, 2020). These animals are traditionally raised in mixed livestock systems and have played an important role in the production of meat and milk, as well as working animals, since their introduction in Brazil at the end of the 19th century

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(ABCB, 2021). Most of the Brazilian buffalo herd is raised in systems of native or cultivated pastures, located in intertropical areas, mainly in the Amazon (GARCIA, 2013). These regions are characterized by high forage yield potential, due to intense solar radiation, high temperatures, and relative humidity.

Legal Amazon is an area delimited by Law 1,806 (January, 6th, 1953) which covers the states of Amazonas, Pará, Acre, Amapá, Rondônia, Roraima, and part of the states of Maranhão, Tocantins and Mato Grosso. In this area, the annual average temperatures vary between 24.3 and 27.1 °C, the maximum temperatures are between 29.5 and 32.0 °C, and the minimum temperatures are between 18.0 to 23.0 °C. Annual rainfall varies between 1,190 mm and 3,000 mm, with the rainiest areas located in the extreme west of the state of Amazonas and the least rainy ones in the southeastern portion of the region (MARTORANO et al., 2018).

Under these conditions, the Amazon region presents a climatic typological pattern that totals 10 climatic subtypes (Af1, Af2, Af3, Am1, Am2, Am3, Am4, Aw3, Aw4, and Aw5), according to Köppen and adapted by MARTORANO et al. (1993) (Figure 1). These climatic subtypes are characterized by monthly average temperatures always above 18 °C and that differ by the volume of rain in the least rainy month. Due to its geographic location, the Brazilian Amazon receives radiation values at the top of the atmosphere that can reach 36.7 MJ $m^{-2}.day^{-1}$ in December-January, and the lowest values of 30.7 MJ $m^{-2}.day^{-1}$ in June-July (SALATI & MARQUES, 1984). Based on the observed meteorological conditions, a climate subdivision of the year into three periods is accepted for the Brazilian Amazon. The rainiest period extends from January to April, with a reduction in the radiant thermal load due to the presence of clouds, whereas the least rainy period goes from August to December, when soil water stocks point out reductions in fodder availability, as a result of periods of water deficits (MARTORANO et al., 2017). The "transition" period comprises the months from May to July, and represents the passage between the end of the rainiest period and the beginning of the least rainy one (BASTOS et al., 2002; MORAES et al., 2005).

Considering the zootechnical activity, the climate is characterized as the main condition of the spontaneous environment, as it acts directly and

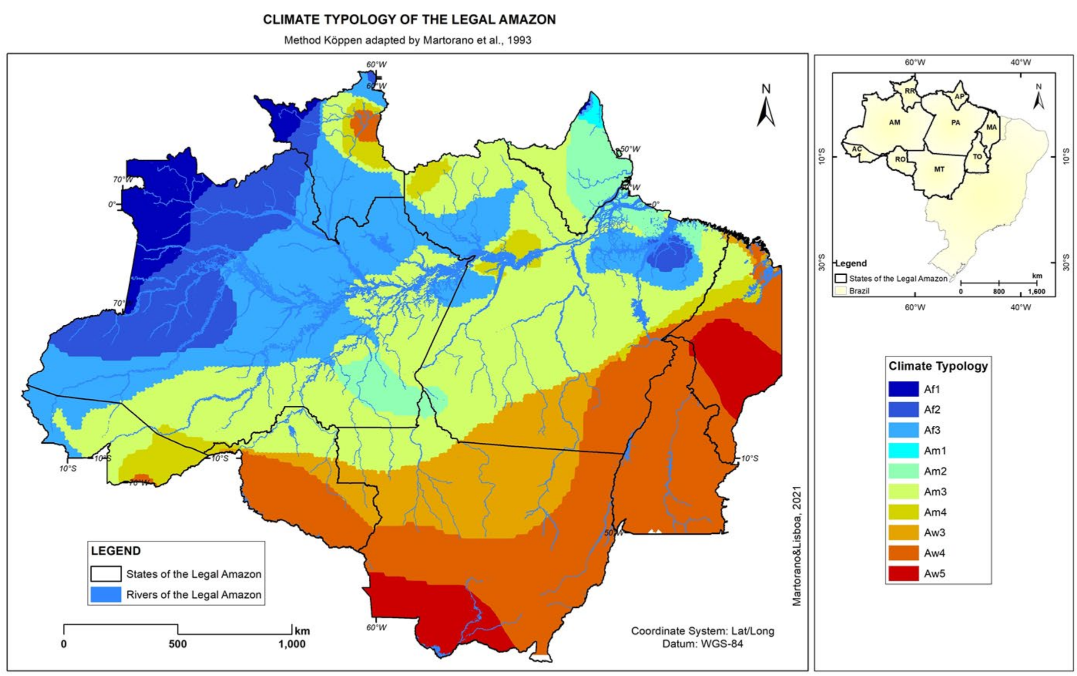

Figure 1 - Map of the climate typology of the Legal Amazon, according to Köppen classification (adapted by MARTORANO et al., 1993).

indirectly on livestock, besides the impossibility of being controlled by the rural producers. Although, water buffaloes are considered animals adaptable to different environments and relatively efficient in dissipating thermal energy (MARAI & HAEEB, 2010a; SILVA et al., 2017), prolonged exposure to tropical climate conditions represents a challenge to these animals. Due to some phenotypic peculiarities, water buffaloes are more susceptible to absorb direct radiation, which can affect their thermoregulatory capacity, leading them to the condition of heat stress (SILVA et al., 2015a; PANTOJA et al., 2018). As thermoregulation in homeothermic animals occurs from passive and active thermolytic mechanisms, the higher frequency of autonomous activation of panting and sweating can impair productive and reproductive performance, due to the higher energy demand for maintaining core body temperature within an ideal range (MISHRA, 2021). In the impossibility of dissipating excess heat, the animals are subjected to a condition of thermal stress.

Physiological changes in heat stress include the reduction in metabolic heat production due to decreased consumption, which reduces the efficiency of feed conversion ratio, considerably impairing the production of meat and milk (SINGH et al., 2012). In environmental conditions of thermal discomfort, animals spend less time feeding and weight gain decreases considerably (GALLOSO-HERNÁNDEZ et al., 2021). Heat stress also reduces milk productivity in buffaloes (CHOUDHARY & SIROHI, 2019) and; consequently, negatively impacts the production of dairy products (LIU et al., 2020). The heat stress is also related to individual factors. As daily milk yield increases, females become more sensitive to heat stress due to the extra metabolic heat associated with additional production (BERNABUCCI et al., 2014). Furthermore, heat stress negatively affects the levels of fat, protein, and lactose in buffalo milk (COSTA et al., 2020).

The fertility of buffaloes is also negatively impacted under heat stress conditions, which reduces the sperm production and seminal quality of bulls kept in the field or in artificial insemination centers (GONÇALVES et al., 2021). This occurs mainly when sires are unable to maintain the average scrotal temperature between 33.0 and 34.0 °C, a range considered normal for adult bulls raised in the Amazon (SILVA et al., 2017). In addition, heat stress has been associated with reduced fertility in buffaloes through its deleterious impact on oocyte maturation and estrous synchronization procedures for artificial insemination (GARCIA, 2006), besides impairing early embryo development, which decreases conception and pregnancy rates (DASH et al., 2016).

Therefore, given the importance of thermal comfort for the productivity of domestic buffaloes, this work gathered and synthesized scientific information on environmental factors that interfere in the buffalo farming, serving as a basis for guiding environmental and animal management measures in tropical climate regions.

Heat stress in water buffaloes

Water buffaloes are homeothermic animals and, therefore, are able to maintain their body temperature within a narrow range, so that it does not vary with the environment temperature (COSTA et al., 2020). To this end, buffaloes use some physiological and behavioral artifices that help to conserve heat, especially when the animal is outside its thermoneutral zone (PORTO et al., 2018; PEREIRA et al., 2020). The thermoneutral zone is defined as the range of ambient temperature considered ideal for maintaining the species in a condition of thermal comfort, a situation in which the need for activation of thermoregulatory mechanisms is minimal, either for heat loss or for thermogenesis. For water buffaloes, the thermoneutral zone is located between 15.5 and 21.2 °C (GOSWAMI & NARAIN, 1962). However, depending on individual adaptability and specific microclimate conditions, buffaloes can withstand higher environmental temperatures without triggering their thermoregulatory mechanisms. For instance, adult bulls were able to maintain their homeothermy when average maximum air temperatures ranged between 31.1 and 31.8 ºC and the average temperature and humidity index ranged from 79.7 to 80.6, in a place with Af climate subgroup (BARROS et al., 2016).

In the Amazon region, where the average temperature is high and the temperature range is small, some buffalo ecotypes may show an increase in body temperature only when the ambient temperature is equal to or higher than 36.0 °C (DANTAS et al., 2008). In fact, animals descended from the first populations of domestic buffaloes that entered Brazil through Marajó Island, in 1890, were naturally selected for adaptation to local climatic conditions, such as high temperatures and humidity, high direct solar radiation, as well as for the food availability initially restricted to forage native to floodplain areas (NASCIMENTO & MOURA CARVALHO, 1993).

Nonetheless, considering the morpho-functional aspects, domestic buffaloes have specific structural particularities, such as a high concentration of melanin in skin and hair (COCKRILL, 1968;

SINGH et al., 2013), reduced density of sweat glands (DEBBARMA et al., 2018), and low hair density (MARAI & HAEEB, 2010a). Although, pigmented skin can minimize the biological hazards caused by ultraviolet radiation, animals with dark skin are more sensitive to heat stress (KOGA et al., 2004; GARCIA, 2013). The exposure of these animals to a thermal environment not included in the thermoneutral range causes instability in the balance between the retention of heat acquired from the environment and/or produced endogenously and the heat effectively dissipated by the individual.

An experiment carried out in the Eastern Amazon from April to August, in the transition between the rainiest and least rainy periods, showed that adult male buffaloes presented mean respiratory rate values of 24.4±3.4 breaths minute^{-1}, heart rate of 64.5±2.7 beats minute^{-1}, and rectal temperature of 38.2±0.5 ºC. Even with a high temperature and humidity index (>80.0), the physiological variables remained within the normal range for domestic buffaloes, indicating that these animals are adaptable, capable of activating their thermoregulatory mechanisms and able to maintain their homeothermy, even under challenging thermal conditions. The mean value of red blood cells, leukocytes, platelets, and hemoglobin did not show significant fluctuations over the months; however, variations were recorded in hematocrit and mean globular hemoglobin (BARROS et al., 2015).

Under heat stress conditions, the hypothalamus synthesizes and secretes corticotropin-releasing hormone (CRH), which reaches the adenohypophysis through the portal system. CRH stimulates the secretion of adrenocorticotropic hormone (ACTH), which, for its part, stimulates the adrenal to secrete glucocorticoids, in which cortisol is the main one, and catecholamines (adrenaline and noradrenaline). The increase in serum cortisol concentration in the acute phase of heat stress is attributed to the intense need for glucose utilization by the animal under these conditions (MÖSTL & PALME, 2002). The increase in the circulation of catecholamines is responsible for immediate responses to stressful situations.

In the case of heat stress, sweating and peripheral vasodilation initially occur, which results in a drop in blood pressure, which is compensated by an increase in heart rate and a subsequent increase in respiratory rate (AGGARWAL & SINGH, 2008; MARAI & HAEEB, 2010b), which generates higher energy demand (GARCIA et al., 2011a) and reduced productivity (WANG et al., 2016). Thus, the need to maintain homeothermy leads to changes in baseline physiological patterns, which may be gauged by measuring variables such as respiratory rate, heart rate, and rectal temperature, making its monitoring useful for the assessment of animal thermal balance (ALVES et al., 2017).

In addition to physiological changes, thermal discomfort causes behavioral changes in animals. One of the outstanding ethological characteristics of the buffalo is the social hierarchy, which is determined through agonistic interactions (DE ROSA et al., 2009). Any situation that increases the animals' reactivity or irritability, such as heat stress, favors the destabilization of the herd's social order. Thermal discomfort also influences the pattern of movement of animals in pastures, which can change the interaction among individuals, in addition to access to food and water (GIRO et al., 2019a).

Behavioral differences were observed in buffalo heifers reared on Marajó Island, Eastern Amazon, when animals kept in an area with natural shading or in a non-forested system were compared. Females without access to shade presented a reduction in the total grazing time, which was concentrated in the intermediate shift (10:00 a.m. to 2:00 p.m.), while buffaloes in wooded areas grazed for a longer time, with distribution of this activity in the afternoon (2:00 p.m. to 6:00 p.m.), early evening (6:00 p.m. to 10:00 p.m.), and early morning (2:00 a.m. to 6:00 a.m.) shifts. Animals in non-forested areas had a destabilization of their idle time, whose normality is fundamental for the psychosomatic repair processes to occur (LI et al., 2021). In turn, female buffaloes raised in the shaded area with higher thermal comfort presented a pattern of behavior more aligned with which is considered normal for buffaloes (ALMEIDA et al., 2019). These results are corroborated by a complementary study, also carried out on Marajó Island, which demonstrated that buffalo females raised in shaded paddocks graze, ruminate, and perform other activities with higher intensity than animals raised without access to shade (ATHAÍDE et al., 2020). In fact, buffaloes raised in silvopastoral systems in tropical regions use the shade of trees to feed, rest, and facilitate heat loss (CASTRO et al., 2008), which includes the habits of wallowing and searching for shade (GALLOSO-HERNÁNDEZ et al., 2020; SANTOS et al., 2021). Lying in the mud in the hottest hours of the day is a common habit of buffaloes raised in northern Australia, where most buffaloes live in the coastal wetlands and river courses, also a region of tropical climate (FAO, 1991).

Physiological variables indicative of the thermal condition in buffaloes

When subjected to threatening climatic conditions, such as high air temperature, high relative humidity, intense direct solar radiation, and heat loads, buffaloes activate physiological thermolysis mechanisms to promote heat dissipation and maintain their homeothermy. However, depending on the intensity of the challenge, buffaloes may present hyperthermia, indicating that these thermolytic mechanisms were insufficient to dissipate the excess heat accumulated (GUDEV et al., 2007a; BROWN-BRANDL, 2018). Thus, because it denotes the result of the thermal energy balance, rectal temperature is considered a more efficient indicator of heat stress than respiratory and heart rate, when considered in isolation (SILVA et al., 2011).

Following a circadian rhythm, the average rectal temperature of buffaloes increases throughout the day, being lower at dawn and higher at dusk (BARROS et al., 2016). The average internal temperature of buffaloes raised in the tropical zone is 38.0 °C in the morning shift, rising to 38.5 °C in the afternoon (SILVA et al., 2017). In tropical conditions, clinically healthy male buffaloes have a mean rectal temperature of 38.2 °C (BARROS et al., 2016). When buffalo calves are raised in a region with a humid tropical climate, internal temperature fluctuations between 38.3 and 39.3 °C are considered normal (MORAES JÚNIOR et al., 2010). Conversely, non-pregnant and non-lactating buffalo cows present an internal temperature ranging from 38.2 to 38.6 °C, when in thermal comfort (SILVA et al., 2011). A similar situation was reported in buffaloes cows raised in pastures with 20% shaded area, a circumstance in which the rectal temperature was 38.6 °C (GARCIA et al., 2011a). It is important to note that the internal temperature of domestic buffaloes can oscillate depending on the age (Table 1) and physiological condition of the animal at the time of evaluation (Table 2 and Table 3).

The maintenance of body temperature aimed to avoid large thermal elevations that may be harmful to the animal's metabolism. In conditions of thermal discomfort, when the internal temperature increases, the respiratory rate also increases, in the animal's attempt to exchange heat by convection, due to the increase in the air flow through the respiratory tract (GIRO et al., 2019a). As, in general, the inspired air temperature is lower than the body core temperature, the contact surface of the respiratory tract cedes heat to the inspired air, leading to heat loss (PEREIRA et al., 2019). Hence, the oscillation of the respiratory rate is a sensitive sign and may precede more significant variations in the rectal temperature (FERREIRA et al., 2006). Also, acceleration of the respiratory rate contributed to heat loss through evaporation, due to the contact of the inspired air with the pulmonary alveoli and the walls of the respiratory ducts.

Table 1 - Internal temperatures of young buffaloes, raised in tropical climate regions, under different management conditions*.

Category	Internal temperature (ºC)	Location (geographic coordinates)	Management condition	Reference
Calves (up to 6 months old)	38.3±0.26 to 39.3±0.36	Belém, PA, Brasil 01°25'S; 48°26'W	silvopastoral system (18-21% of shaded area)	MORAES JÚNIOR et al. (2010)
Calves (6 months old)	39.14±0.07 to 40.0±0.10	Chaitnat Province, Thailand 15°16'N; 100°06'E	free stall (modified roof or normal corrugated iron roof)	KHONGDEE et al. (2013)
Young males (7 to 9 months old)	38.5±0.37 to 40.5±0.10	Karnal, Índia 29°70'N; 76°98'E	open stable, no shading	DAS et al. (1999)
Young males (9 to 12 months old)	38.6±0.30 to 38.9±0.20	Dehong State, China 24°43'N; 98°57'E	shaded or non-shaded barns	GU et al. (2016)
Young males (1 to 2 years old)	38.4	Karnal, India 29°70'N; 76°98'E	closed stable	HAQUE et al. (2012)
Steers (4.5 years old)	38.2±0.5	Castanhal, PA, Brazil 1°18′18″S; 47°56′36″W	confined in collective stalls	BARROS et al. (2015)
Heifers (1.5 years old)	38.36 to 38.88 38.16 to 39.32	Pirassununga, SP, Brasil 21° 57'S, 47°27W	outside of the climatic chamber simulated heat wave	PEREIRA et al. (2020)

*Adapted from GARCIA (2013).

Table 2 - Internal temperatures of adult female buffaloes, raised in tropical climate regions, under different management conditions*.

Category	Internal temperature (ºC)	Location (geographic coordinates)	Management condition	Reference
Non-lactating and non-pregnant adult females (2.5 years old)	39.30±0.37	Los Banos, Filipinas 14°15'N; 121°24'E	confined in individual stalls	KOGA et al. (2004)
Adult females in early lactation stage	38.4±0.1 38.0±0.1	Karnal, India 29°42'N, 76°58'E	access to sprinkler showers access to water for bath	AGGARWAl & SINGH (2008)
Non-lactating and non-pregnant adult females (4 to 5 years old)	38.2±0.1 to 38.8±0.1	Belém, PA, Brazil 01°26'S; 48°26'W	pasture, with neither shading nor water for bath	SILVA et al. (2011b)
Non-lactating and non-pregnant adult females (4 to 5 years old)	38.2±0.1 to 38.6±0.1	Belém, PA, Brazil 01°26'S; 48°26'W	silvopastoral system	SILVA et al. (2011b)
Non-lactating and non-pregnant adult females (6 to 7 years old)	38.94±0.56 to 39.11±0.48	Belém, PA, Brazil 01°25'S; 48°26'W	pasture, no shading, with water for bath	GARCIA et al. (2011b)
Non-lactating and non-pregnant adult females (6 to 7 years old)	38.62±0.48 to 38.68±0.32	Belém, PA, Brazil 01°25'S; 48°26'W	silvopastoral system (20% shaded area)	GARCIA et al. (2011b)
Lactating adult females (not reported age)	38.66 38.25	Pattoki, Pakistan 31°02'N; 73°84'E	stall with shade stall with shade, fans and sprinklers	AHMAD et al. (2019)

*Adapted from GARCIA (2013).

In tropical climate conditions, the respiratory rate of water buffaloes at rest is 18 mov min^{-1}, reaching 24 mov min^{-1} in the hottest periods of the day. As buffaloes have a low relative efficiency in heat loss through the skin, loss through expired air, characterized by an increase in respiratory rate, plays an important role when compared to sweating (GUDEV et al., 2007b; JOSET et al., 2018). In the Brazilian Amazon, buffalo bulls managed in an artificial insemination center had a respiratory rate of 21 to 23 mov min^{-1}, in the rainiest period of the year, increasing to 23 to 30 mov min^{-1}, recorded in the transition period (BARROS et al., 2015).

With the activation of thermoregulatory mechanisms in thermal stress, there is an increase in heart rate (MARAI & HAEEB, 2010b). Female buffaloes raised in the Amazon present a lower heart rate in the morning than in the afternoon, either in the rainiest period of the year (61.1 *vs* 65.5 bpm), in the transition period (54.5 *vs* 63.2 bpm), or in the least rainy period (55.5 *vs* 58.1 bpm) (SILVA et al., 2011). The increase in heart rate observed in the rainiest period of the year is related to the increase in the relative humidity, characteristic of times with higher rainfall (MARAI et al., 2009). A higher relative air humidity means it is less easy to vaporize a given mass of fluid, such as sweat, for example, which leads to a smaller amount of heat lost per unit of time (PEREIRA et al., 2019). Conversely, the increase in heart rate in the afternoon period basically happens as an immediate response to the increase in air temperature, whose maximum values occur between 12pm and 3pm in tropical climate regions.

The temperature of the animals' body surface is directly influenced by the thermal environment, which affects thermal exchanges through the skin surface (GIRO et al., 2019a). For the maintenance of homeothermy, the skin plays an important role, as heat loss occurs through transpiration (PEREIRA et al., 2020). This loss depends on the temperature gradient between the animals' body surface and the microenvironment that surrounds them. Thus, the assessment of surface temperature provides relevant information about the animal thermal balance and it is an easily obtainable measure, associated with rectal temperature and

Table 3 - Internal temperatures of adult male buffaloes, raised in tropical climate regions, under different management conditions*.

Category	Internal temperature (ºC)	Location (geographic coordinates)	Management condition	Reference
Adult males (3 to 4 years old)	37.20	Karnal, India 29°70'N; 76°98'E	closed stable	HAQUE et al. (2012)
Adult males (4 to 5 years old)	37.9±0.1 to 38.7±0.1	Castanhal, PA, Brasil 01°18′S, 47°56′W	open stable, with partial shading	SILVA et al. 2018
Adult males (7 years old)	37.5 to 38.6	Viseu, PA, Brasil 01°11' S, 46°08'W	open stable, with partial shading	GONÇALVES et al. (2021)

*Adapted from GARCIA (2013).

respiratory rate (SALLES et al., 2016). The ideal for domestic animals is that there is a gradient of 6 °C + 6 °C between the temperature of the body core and the skin surface and between the skin surface and the air, so that the accumulated thermal energy can flow centrifugally and without causing overheating in the animal's body (MEDEIROS & VIEIRA, 1997).

Since body surface temperature is correlated with climatic variables, especially air temperature, relative humidity, and global solar radiation, its assessment carried out strategically at certain periods of the day provides relevant information on thermal comfort (GIRO et al., 2019b). In the Amazon, it was reported that female buffaloes have higher surface temperature values in the afternoon and in the least rainy period of the year, and this temperature has a positive correlation with the ambient temperature (0.52) and a negative correlation with the relative humidity of the air (r = -0.39). (SILVA et al., 2011). Like internal temperature, buffalo body surface temperature is lower in the morning than in the afternoon, regardless of whether it is measured on the right flank (31.9 ± 2.1 *vs* 35.2 ± 1.6 ºC) or on the left flank (32.4 ± 2.0 *vs* 35.4 ± 1.7 ºC) (BARROS et al., 2016).

Climatic variables interfering with thermal comfort

In order to understand how climatic variables affect the thermal comfort of buffaloes in the Amazon, it is necessary to know that two distinct management systems are adopted in the region. The first is an extensive management mode that uses traditional production areas in which buffaloes are raised on native pasture ecosystems on dryland and on periodically flooded land along the Amazon River and tributaries. It occurs mostly in the Marajó Island on the Amazon mouth, the banks of the river and its tributaries, and in the states of Amapá and Roraima. The other management mode is conducted in areas of intense cattle ranching where cultivated pastures have been occupying previously anthropized areas and where herds have more productive animals. In these areas, initiatives to adopt silvopastoral systems have been promoted, based on planted forests and cultivated pastures (SILVA et al., 2021). Both dryland and floodplain areas in the Brazilian Amazon are under the influence of the tropical climate, predisposing animals to its effects.

One of the climatic variables with the highest influence on the environment in which the animal is found is air temperature, which, in tropical regions, is high practically throughout the year (GARCIA, 2013). Buffaloes exposed to ambient temperatures of 36 °C or higher, high air humidity, and direct solar radiation present increased internal temperature, respiratory rate, and heart rate, even when they are able to eliminate part of the excess body heat through skin evaporation (DANTAS et al., 2008). The exposure of animals to high ambient temperatures promotes an increase in blood flow from the central core to peripheral regions of the body (Figure 2). This autonomous response occurs in the animal's attempt to eliminate accumulated internal heat, contributing to the increase in surface temperature (SILVA et al., 2011). Experimentally, it was observed that the air temperature in a bioclimatic chamber from 30.9 to 36.0 °C causes an increase in respiratory rate and panting in buffaloes, reaching 62.5 mov min^{-1} (GUIMARÃES et al., 2001).

Relative humidity is another variable that influences the caloric balance in hot environments, where heat loss through evaporation is crucial for the maintenance of homeothermy. In environments for raising domestic animals, the ideal relative humidity should be from 50 to 70% (BAÊTA & SOUZA, 2010). High and low air humidity are

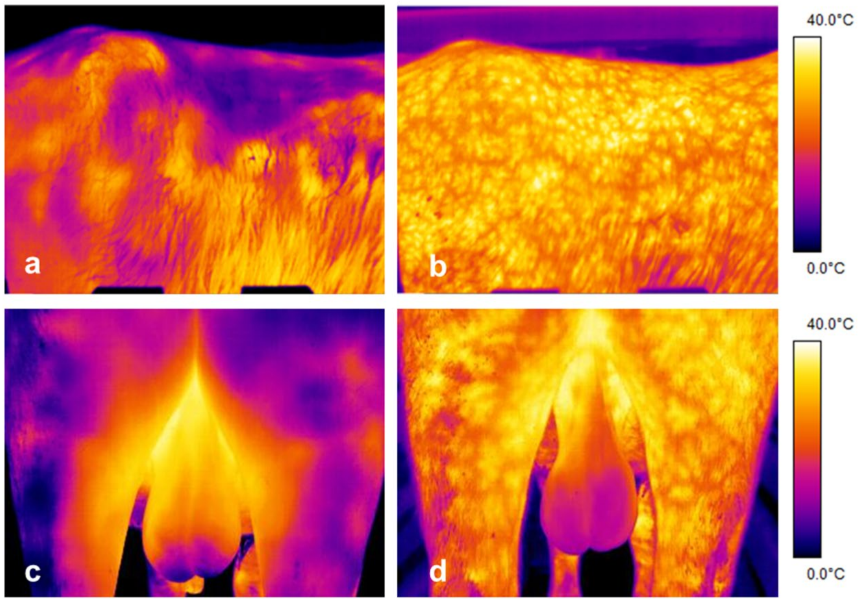

Figure 2 - Illustrative thermographic images acquired in the right lateral (a, b) and caudal (c, d) positions of water buffaloes raised in Eastern Amazon, recorded on the same assessment day in the morning (a, c) and afternoon (b, d) periods. It is worth highlighting the change in the skin thermographic pattern, due to peripheral vasodilation that occurred in the hottest part of the day, in order to facilitate the loss of body heat (Source: GARCIA, A.R., personal archive).

equally harmful to animal health when associated with high air temperature. If the environment is hot and very dry, evaporation is rapid, causing cutaneous irritation and general dehydration. It may occur in hot and arid regions or in specific situations of heat stress in a tropical climate. In hot and very humid microclimate conditions, evaporation is slow or null, reducing thermolysis and increasing the retention of thermal energy, as a result of damage to heat loss by convection in this condition (SILANIKOVE, 2000).

Depending on the intensity and duration of exposure to direct solar radiation, animals may show changes in certain physiological variables. Tolerance to direct solar radiation occurs due to physical characteristics, such as fur and skin color, which in buffaloes, because it is dark, makes them more sensitive (GARCIA, 2013). Solar radiation also interferes in animals' surface temperature, increasing its value and changing the thermal gradients between the body central and superficial core, the surface, and the environment, hampering the thermoregulatory process (GIRO et al., 2019a).

Studies carried out with buffaloes raised in the Amazon indicate the following variation as a normality standard for blood parameters: red blood cells (4.7-11.8 x10^6 μl^{-1}), leukocytes (5.4-15.4 x10^3 μl^{-1}), platelets (101-775 x10^3 μl^{-1}), hemoglobin (8-16 g dL^{-1}), hematocrit (25-45 %), mean globular volume (37-66 fL), globular hemoglobin (11-19 pg), and mean globular hemoglobin concentration (27-36 g dL^{-1}) (FONTES et al., 2014). Cortisol concentration ranges between 1.7±0.2 and 1.9 ± 0.3 mg dL^{-1}, with higher values recorded when animals are raised without access to shading (SILVA et al., 2014), being a valid bioindicator of stress condition, as long as its circadian pattern of release is considered in the planning of sample collections. For their part, the concentrations of triiodothyronine (T3) and thyroxine (T4) of dairy buffaloes raised in the Amazon were negatively correlated with air temperature and with BGHI, indicating that the serum concentration of thyroid hormones decreases as the biophysical indicators of heat stress increase (SILVA et al., 2014).

Animal thermal comfort indexes of interest for tropical buffalo farming

Thermal comfort indexes have been commonly used to better assess the impact of environmental thermal conditions on animals. In the Eastern Amazon, several thermal comfort indexes were positively correlated with internal temperature, body surface temperature, and respiratory rate, demonstrating that they may be useful to indicate favorable environments to cause heat stress in animals. Among these indexes, it is possible to mention some that are historically used for animal monitoring in hot climates, such as the Temperature Humidity Index (THI) (THOM, 1959), the Black Globe Humidity Index (BGHI) (BUFFINGTON et al., 1981), the Radiant Heat Load (RHL) (BOND & KELLY, 1955), and the Benezra Comfort Index (BCI) (BENEZRA, 1954). It is also worth pointing out new indexes that have been developed with the aim of increasing the specificity of measuring heat stress in water buffaloes raised in the Amazonian environment.

THI is calculated by associating air temperature and relative humidity, constituting a fair environmental indicator, which may be used for agricultural purposes and for estimating animal thermal comfort (NASR, 2017). In order to determine the thermal comfort of buffaloes, THI values ≤74 are considered more adequate (SOMPARN et al., 2004), so that higher values could indicate discomfort or heat stress. However, studies carried out with buffaloes in tropical highland regions indicated that the thermal discomfort would be higher when THI reaches 85 (ABLAS et al., 2007). For calves raised in a humid tropical climate region, the THI between 78 and 79 did not alter the physiological parameters (MORAES JÚNIOR et al., 2010). It is also known that with a THI between 74.3 and 78.4, buffalo bulls may show no sign of thermal discomfort when protected from direct solar radiation. However, the THI showed a significant negative correlation with the integrity of the plasma membrane of buffalo bulls' spermatozoa (-0.17), indicating that more severe environmental conditions can reduce the quality of the ejaculate (BARROS et al., 2015). The THI also demonstrated that there is an association between climatic conditions and the productive performance of lactating buffaloes raised on the Asian continent, with a reduction in daily milk production and 1% losses in productivity for each elevation unit when the THI exceeds 82 (CHOUDHARY & SIROHI, 2019).

BGHI assesses the thermal comfort condition of animals and takes into account measurements of black globe temperature, dew point temperature, and ambient temperature (BUFFINGTON et al., 1981). BGHI has been successfully used to evaluate the thermal comfort of cattle raised in pasture production systems (GIRO et al., 2019a). BGHI is also a dimensionless index, which presents the following classification: up to 74, it indicates thermal comfort condition; from 74 to 78, alert condition; from 79 to 84, dangerous condition; and above 84, emergency condition (SOUZA et al., 2002). It is a versatile indicator, as it directly assesses the effects of radiation, air temperature, and relative humidity and, indirectly, the effects of ventilation on the environment where the animals are kept (LI et al., 2009). Studies carried out with buffalo cows demonstrated that BGHI is correlated with physiological variables that are relevant to the thermolysis process, such as respiratory rate ($r = 0.48$; $P < 0.05$), heart rate ($r = 0.30$; $P < 0.05$), body surface temperature ($r = 0.90$; $P < 0.05$), and internal body temperature ($r = 0.59$; $P < 0.05$) (SILVA et al., 2011).

RHL is the indicator that shows the total radiation received by a body from all surrounding space. This definition does not encompass the net exchange of radiation between the body and its surroundings, but includes the radiation incident on the body (BOND & KELLY, 1955). Solar radiation is the main source of environmental heat acquired by animals (SAMPAIO et al., 2018). Higher THI values (81.8 ± 2.6 *vs* 77.7 ± 2.6) and rectal temperature (38.4 ± 0.4 *vs* 38.0 ± 0.5) recorded in the afternoon are closely related to higher radiant heat load received by the animals during this period of the day (BARROS et al., 2016).

BCI was specifically developed to assess the thermal comfort of animals raised in tropical climate conditions. Its mathematical formula includes two relevant physiological variables: rectal temperature and respiratory rate. For BCI values close to two (2.0), it is considered that animals have a higher degree of adaptability to the tropical environment and, for values higher than two, animals have adaptive difficulties (BENEZRA, 1954). For buffalo calves, the mean BCI of 2.9 does not indicate physical and behavioral characteristics compatible with heat stress, as their respiratory frequency is physiologically higher than that of adult animals (MORAES JÚNIOR et al., 2010). In the Amazon, buffalo heifers had higher BCI in the morning period (2.2), probably associated with lower air temperature and solar radiation compared to the afternoon (2.4) (SILVA et al., 2010). Because it is an indicator calculated by monitoring physiological variables, BCI is highly correlated with respiratory (0.97) and heart rate (0.89) (BARROS et al., 2015).

Considering that the classical thermal comfort indexes were invariably validated, originally, in conditions very different from those occurring in the Amazon region, more recent studies have determined and validated new thermal comfort indexes for buffaloes kept in the environmental conditions of the Eastern Amazon. Thus, there was the development of indexes that were premised on modeling and adjusting the joint effect of the climatic variables of air temperature, relative humidity, dew point temperature, wet bulb temperature, and black globe temperature on the physiological variables of body surface temperature, respiratory rate, and rectal temperature. Among these alternative indicators, the Buffalo Comfort Climate Condition Index (BCCCI) and the Buffalo Environmental Comfort Index (BECI) showed a high rate of agreement and significant correlation with physiological and climatic variables (SILVA et al., 2015). For this reason, these indicators are recommended to diagnose the heat stress condition of buffaloes raised in the Amazon and, due to their easy applicability, they may allow management interventions in order to increase thermal comfort and, consequently, these animal's productivity (PANTOJA et al., 2018).

Technologies to identify thermal stress in water buffaloes

Climate change is noticeable around the world and the frequency of heat loads has gradually increased (SAVALIYA et al., 2019). Since heat stress impairs animal productivity, the use of technological devices to monitor the environment and the animals' responses to heat challenge can help increase the efficiency of the activity and contribute to animal comfort and welfare. Therefore, the adoption of precision livestock farming instruments has gained importance in research and in rural properties dedicated to agri-food production, which includes buffalo farming.

Within this concept, computers, video cameras, thermal cameras, wearable or non-wearable sensors, among others, have been used to monitor animals, autonomously or inductively, in order to identify physiological and/or behavioral changes. For the automation of a production system to take place, the first-layer technology is the electronic individual identification of animals, which is crucial for the aggregation of other technologies. For buffaloes, the use of subcutaneous transponders that operate in radio frequency (RFID) has been shown to be an efficient, safe, and accurate way of electronic identification, whether for adult or young animals. This technology can be adopted for animals raised in dryland or floodplain pastures, common in the Amazon environment, with an efficiency of 93.9% and 97.2%, respectively (GARCIA et al., 2020).

After electronic identification, the thermal condition of buffaloes can be monitored through non-invasive methods such as infrared thermography (BARROS et al, 2016; BRCKO et al., 2020), because buffaloes in hot conditions increase blood volume and flow to the skin surface in order to facilitate heat dissipation (KOGA, 1999). The use of infrared thermography allows for accurate measurements of body surface temperatures at distances that can vary from 0.3 to 5.0 meters, depending on the technical characteristics of the equipment used. Since infrared thermography uses sensors capable of identifying thermal oscillations on body surface and in the rearing environment, the technique has shown increasing utility in Veterinary Medicine and Animal Science (CHACUR et al., 2017; MENDOZA et al., 2020).

Its utility has been proven in the evaluation of thermal comfort of buffalo bulls raised in an Amazonian environment, due to the correlation observed between the surface temperatures of the ocular globe and flank region with the internal body temperature (BARROS et al., 2016). Furthermore, infrared thermography allowed the identification of the normal scrotal thermoregulatory pattern of buffalo bulls raised in the Amazon, characterized by three parallel thermal bands, with evident symmetry between right and left testicles (SILVA et al., 2017). The technique also allowed the identification of significant negative correlations between the temperatures of ocular globe, epididymis, and scrotum with sperm volume and progressive sperm motility ($P < 0.01$) (SILVA et al., 2017).

Infrared thermography can also be used to assess the thermoregulatory responses of female buffaloes raised in a hot and humid climate. Higher surface temperatures are observed in dairy females kept on pasture in the Amazon, between 12 p.m. and 6 p.m., with lower body temperatures recorded at 9 p.m. Moreover, ocular globe and cheek temperatures in female buffaloes have a positive correlation with core body temperature and respiratory rate (BRCKO et al., 2020). The assessment of surface temperature through infrared thermography of anatomical regions such as the mammary gland can broaden the understanding of the microvascular mechanisms of this organ, providing additional information on mammary functioning (MENDOZA et al., 2020).

In addition to aspects intrinsic to animals, infrared thermography used in distal sensing can help identify thermal environmental conditions, such as pasture, soil, and forestry components of integrated production systems in tropical climate, elements that directly influence animals' thermal comfort and productive efficiency. Serial thermography identifies the thermal rise in the environment, which causes an increase in water vapor output demands, both from pastures and from the soil, causing a reduction in water availability to plants, which compromises the development of pastures and reduces productivity in livestock (PILATO et al., 2018).

Given that environmental characteristics can cause changes in animal behavior (SILANIKOVE, 2000), some behavioral responses can be indicators of heat stress and welfare. Among these, the number of agonistic interactions per unit of time, excessive movement or idleness, changes in rumination and displacement patterns, and frequency of access to drinking fountains are mentioned (OLIVEIRA & RANGEL, 2013; GARCIA et al., 2019; GIRO et al, 2019a). Therefore, the use of wearable sensors has been useful in animal monitoring. Accelerometers can be attached to collars and provide an electrical signal, which is sent to software that records and interprets changes in the animals' behavior. Another technology of interest is the use of electronic ear tags containing microchips that store information about the animal and its location, which are capable of monitoring the position of the herd, sending the data to a control and management software. Thus, with the advance of technological development, it is noticeable that more devices will be available for use in livestock in the near future, generating increasingly robust databases and greater processing capacity, which will be useful to guide decision-making and raise the productive indexes of the herds.

Strategies for mitigation of heat stress in buffaloes

As animal productivity and welfare are directly affected by the tropical climate (MADER et al., 2010; MARAI & HAEEB, 2010a; STORTI et al., 2019), strategies that reduce these negative impacts become essential. In a first approach, the identification and selection of animals or breeds that are more adapted and thermotolerant is a promising strategy. This selection trend may be driven by recent climate changes (HOFFMANN, 2010; ALVES et al., 2017), as increasingly frequent heat loads have also affected buffalo populations raised in temperate zones and which, until then, remained exempt from damage caused by heat stress.

It is known that among the populations of different buffalo genotypes used for meat, milk and labor production, there are more adaptable individuals with a more efficient thermoregulatory system. These ones stand out for their individual acclimatization capacity and higher plasticity, and are able to quickly compensate for caloric gains, dissipating thermal energy and preserving their homeothermy. Animals with these characteristics are able to maintain physiological, hematological, and seminal parameters at normal levels, may be identified and, consequently, used in the selection of buffaloes with higher tolerance to heat (BARROS et al., 2015; SILVA et al., 2017). Not by chance, the heat tolerance tests developed in Brazil, first for buffaloes, and later adapted for beef and dairy cattle, demonstrated high efficacy in discriminating more adaptable individuals and more capable of responding positively to the challenges of natural irradiation and recovery at relatively short intervals. Thus, the identification of genes associated with heat tolerance, their incorporation into genetic improvement programs, and the inclusion of the effects of biometeorological covariates in selection indexes should be targeted for genetic evaluation of animals raised in hot climates.

Parallel to the identification of bulls and dams that are more adaptable to the tropical climate, it is also possible to directly intervene in some components that are part of the architecture of production systems, aiming to increase the buffaloes' thermal comfort. The availability of water for immersion facilitates heat dissipation, as heat stress increases the volume and blood flow to the peripheral vessels of the skin surface, raising the skin temperature and allowing heat exchange between animal and environment. By providing shade and water for bathing to buffalo cows raised on pasture, it is noticed that they have a predilection for the act of immersion when they are in conditions of extreme thermal discomfort, being able to practice it even during rumination periods (ABLAS et al., 2007).

However, water is a scarce natural resource and agricultural activity is recognized for its large water demand (DOREAU et al., 2012). In this sense, the use of water intended for the immersion of animals, in artificial or natural enclosures, has been oriented towards reduction. Therefore, the availability of shaded areas for animals is currently the most sustainable and effective method to reduce heat stress on animals. Shading reduces the impact of solar radiation on animals, especially in tropical regions, where this radiation amplifies the negative effects of high temperature environments (SILVA et al., 2014). The shade offered to the animals may be natural or artificial, and the choice of the model has great economic and biological relevance (GARCIA, 2013), as it is reported that cows prefer to take shelter in shades with a higher degree of blocking of direct solar radiation (SCHÜTZ et al., 2009).

Mobile artificial shading, a management strategy in which polypropylene screens are generally used, produces less protection against solar radiation compared to fixed shading, which is made with fiber cement or galvanized tiles (SHOSHANI & HETZRONI, 2013). Even so, a screen is an alternative that may be resorted to when immediate effects are required to cool the animals down. Buffaloes kept in screened sheds have lower internal temperature (39.1 ± 0.1 *vs.* 40.0 ± 0.1 °C; $P < 0.01$) and lower water consumption (29.7 ± 0.8 *vs.* 34.1 ± 1.1 L animal^{-1} day^{-1}; $P < 0.01$) when compared to buffaloes raised without access to screened shelters (KHONGDEE et al., 2012). Still, the choice of places to be shaded within the production system differently modulates the response of animals to the offer of shade, which is more effective as the shading reaches larger areas of permanence of the animals. The use of shading in the waiting room for milking, for instance, is a strategy that can reduce, to some degree, the heat stress of lactating animals (CERUTTI et al., 2013).

Despite requiring longer periods to check results, the implantation of the arboreal component has brought very consistent results for animal comfort. The natural shading of pastures is able to favor not only the thermal comfort of female buffaloes, but also their milk production and the performance of the calves until weaning (MORAES JÚNIOR et al., 2010). In addition to mitigating the effects of the environment on buffaloes, the use of pastures integrated with forest production is ecologically recommended and favors the enrichment of the environment (GARCIA 2013; JOSET et al., 2018). In the Amazon region, buffaloes have a lower internal temperature when they have access to natural shade (38.6 ºC) compared to those raised without shading (38.8 ºC) (SILVA et al., 2010). This fact is evidenced in the afternoon period compared to the morning, regardless of the period of the year (SILVA et al., 2011). It is considered that, for buffaloes, adherence to environmental management practices, such as the use of silvopastoral systems, represents an efficient alternative to increase thermal comfort and animal welfare (SANTOS et al., 2020; MORAES JÚNIOR et al., 2010). In this type of system, there is an increase in productivity due to the improvement in the feed conversion rate, caused by the milder microclimate resulting from tree planting (CASTRO et al., 2008). In silvopastoral systems, the canopies reduce the incidence of solar radiation on animals, allowing the temperature to be reduced between 2 and 3 °C when compared to full sun exposure (LOPES et al., 2016), providing higher thermal comfort to animals (LEMES et al., 2021; FAÇANHA et al., 2013).

In addition to dams and calves, the provision of natural shade also favors buffalo bulls in the Amazon, meaning that the seminal quality does not present significant changes, even in times of the year with hotter temperatures and more intense solar radiation (BARROS et al., 2015). One of the possible explanations for the beneficial effect of natural shading on buffalo bulls is the reduction of BGHI in forested systems (GARCIA, 2013), which favors the animal's body thermoregulation and, consequently, reduced the incidence of testicular degeneration. This is a pathological condition that may be caused by failure in scrotal thermoregulation and drastically reduces the seminal quality of buffaloes by increasing the incidence of sperm defects (GONÇALVES et al., 2021).

The implantation of silvopastoral systems in buffalo farming in the Eastern Amazon also avoids energy expenditure for thermoregulation and promotes increase productivity (CASTRO et al., 2008; GARCIA et al., 2010). Female buffaloes raised in silvopastoral systems show a reduction in heart rate and internal temperature (GARCIA et al., 2011a). Female buffaloes with access to arboreal shading present a better condition of comfort and, therefore, a reduction in cortisol levels and an increase in serum triiodothyronine levels, which demonstrated the efficiency of the silvopastoral system in minimizing heat stress, especially in the afternoon (SILVA et al., 2011). A higher productivity of meat and milk was observed in silvopastoral systems when compared to traditional systems, which is directly related to the comfort environment, with shading made available to buffaloes (GALLOSO-HERNÁNDEZ et al., 2021). Buffaloes raised in silvopastoral systems also produced a higher percentage of prime cuts, and meat with better texture and tenderness (JOELE et al., 2013).

Finally, within the context of mitigating the impacts of tropical climate effects and heat loads on animals, the use of precision livestock farming elements can be added. Precision livestock farming is a set of processes that allows the collection of data from animals and the surrounding environment in real time, allowing for their interpretation and instantaneous decision-making, which can greatly benefit production animals. Among its applications, some have been developed to control parameters that are directly related to heat stress, such as monitoring respiratory rate (CARVALHO et al., 2020), internal temperature (SHARPE et al., 2021), surface temperature (GIRO et al., 2019b; ROMANELLO et al., 2018; BARROS et al., 2015), and animal's grazing behavior. Furthermore, a range of different devices has been used for rigorous real-time monitoring of the environment where the animals are kept, even when raised on pasture (PEZZOPANE et al., 2019). Hence, data collected and analyzed through the use of increasingly elaborated predictive mathematical models regarding animal comfort indexes have helped to implement adequate strategies to mitigate animal heat stress (PACHECO et al., 2020; BROWN-BRANDL, 2018; MADER et al., 2010).

CONCLUSION

Despite their relative adaptability to adverse environmental conditions, water buffaloes suffer from heat stress, especially when exposed to direct solar radiation in the Amazon. Therefore, knowledge about the factors that affect buffaloes' ambience when raised in a tropical environment, as well as about the management measures that provide higher thermal comfort, are essential. Furthermore, the selection of thermotolerant animals is an important step in the buffalo production in the tropics and should be stimulated by modern phenotyping and genotyping mechanisms. At the same time, the technological development of systems that incorporate the arboreal component to tropical pasture areas has shown that integrated systems are an excellent option to provide ambience and higher thermal comfort to beef and milk buffaloes. Finally, the advent of precision livestock farming and the improvement of algorithms that consider the specific responses of buffaloes in tropical production systems may contribute to the development of processes for monitoring the condition of animals in real time and immediate decision-making, aiming at raising productivity in association with animal welfare.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; and in the decision to publish the results.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. The authors critically revised the manuscript and approved of the final.

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