Mitigation Strategies for Heat Stress on Dairy Buffalo

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Increases in temperature and the greater incidence of extreme events are the consequences of the climate change that is taking place on planet Earth. High temperatures create severe discomfort to animal farms as they are unable to efficiently dissipate their body heat, and for this, they implement mechanisms to reduce the production of endogenous heat (reducing feed intake and production). In tropical and subtropical countries, where buffalo breeding is more widespread, there are strong negative consequences of heat stress (HS) on the production and quality of milk, reproduction, and health. The increase in ambient temperature is also affecting temperate countries in which buffalo farms are starting to highlight problems due to HS.

dairy buffalo heat stress feeding milk yield and composition

1. Heat Stress Introduction

The demand for livestock products is growing rapidly, driven by population and income growth plus urbanization. Simultaneously, livestock production is facing increasing pressure from climate change effects, such as increasing temperatures, more variable precipitation patterns, more frequent extreme events, and increasing carbon dioxide concentration [1]. The global temperature has increased by 1.5 °C above pre-industrial levels [2]. Moreover, the increasing temperature is accompanied by water scarcity, and this condition leads to a more severe situation of alert [3]. Given the current climate scenario and not very encouraging future estimates [4][5], we must expect farming to be increasingly adversely affected by the effects of HS. Heat stress can be simply defined as a condition that occurs when an animal cannot dissipate an adequate quantity of endogenous heat, whether it is produced or absorbed by the body, to maintain body thermal balance [6]. This can happen when the ambient temperature and humidity are high and can cause a range of negative effects on the animal's health and productivity. Thermal stress is triggered when environmental conditions exceed the upper or lower critical temperature of domestic animals requiring an increase in basal metabolism to deal with the stress [7]. Heat stress occurs when the ambient temperature is above the thermoneutral zone.

It is well known that HS can cause a range of negative effects on dairy cattle, including decreased milk production, reduced feed intake, reduced reproductive performance, and increased susceptibility to diseases [8]. In addition, HS can cause physiological changes such as an increased heart and respiration rate, increased blood flow to the skin, and increased sweating which can lead to a loss of electrolytes, dehydration, and inflammation. For these reasons, HS has a huge economic impact on the global dairy industry.

Little information is available on the effect of HS in buffalo reared under intensive conditions. Buffalos are widespread in many areas of the world, mainly in Asia, some Mediterranean countries, and some Eastern European and Latin American countries [9]. Therefore, buffalos are adapted to different climates. In particular, due to their morphological, anatomical, and behavioral characteristics, buffalos are better adapted to hot and humid climates [9]. Compared to cows, it appears that buffalos are more tolerant to HS and can better handle tropical climates, with fewer negative effects on production and physiology. However, they exhibit signs of greater distress when exposed to direct solar radiation [9][10]. The body temperature of buffalo is slightly lower than that of cattle, but buffalo skin is usually black, so their body absorbs a large amount of solar radiation due to their dark skin and sparse hair, and, in addition, buffalo have a less efficient evaporative cooling system due to their rather poor sweating capacity compared with cattle [9]. In fact, buffalo skin has one-sixth of the density of sweat glands compared to cows; thus, buffalo dissipate heat poorly by sweating [9]. The maximum number of sweat glands/mm^2 on buffalo skin was observed the in head dorsal followed by the abdomen dorsal, tail dorsal, neck dorsal, and thorax dorsal areas [11]. The distribution of sweat glands, the dark color of the skin, and the sparse body hair adversely affect heat tolerance in these animals.

The greater presence of buffalo is mostly found at latitudes characterized by a hot–humid climate (tropical and subtropical areas) and for this reason, they have developed defensive mechanisms over time such as a large quantity of melanin and sebaceous glands which, respectively, prevent ultraviolet rays from being able to pass through the dermis [12] and, through the sebum secreted and dissolved by high temperatures, reflect the sun's rays [13]. These glands secrete sebum, a fatty substance that covers the surface of the skin and coats it with a lubricant, making it slippery to water and mud. This fatty sebum, together with the thick upper horny layer of the skin, prevents the water and solutes it contains from being absorbed by the skin. In this way, the animal is protected from the harmful effects of any deleterious chemical compounds present in the water. Furthermore, the sebum layer melts during the hot season and becomes more shiny reflecting many of the Sun's rays, thus relieving the animal from excessive external thermal load.

Buffalos are more heat-stressed (HSed) when they are prevented from displaying their adaptive behavioral traits such as seeking shelter, wallowing, and/or diving into water [14]. It has been shown that buffalos are better able to regulate their body temperature if left to splash around in pools of water rather than using showers as a cooling system [15]. It is not always possible to have pools; however, like any other animal, they still need to have appropriate measures for HS management to maintain their productivity and health.

The exposure of buffalo to hot conditions causes a series of changes in their biological functions which includes a decrease in feed intake, the efficiency and utilization of their diet, disturbances in the metabolism of water, proteins, energy and mineral balances, enzymatic reactions, hormonal secretions, and blood metabolites [16], a reduction in milk yield and quality [17], and a reduction in the manifestation of heat and therefore fertility [18]. The effects of HS on the quality of production are less evident than in cattle [17]; the negative impacts of HS on various biological functions are discussed in more detail in the source paper.

High air temperature and humidity are the main factors responsible for variations in the physiological reactions of animals, and these variations can be different depending on the species and breed [8][19]. As reported in the literature [20], thermal parameters are the key factors used to calculate heat transfer: these include temperature, humidity, wind speed, and solar radiation. The combination of those parameters results in several indicators that are useful to determine the stress condition to which animals are exposed. The most widely used is the temperature–humidity index (THI), which combines air temperature with humidity providing a single index that gives information on animal comfort. The temperature–humidity index is mostly used for the measurement of HS condition because data relating to other parameters that could affect HS, such as solar radiation, wind speed, and precipitation, are not always available.

Numerous studies have established THI thresholds for HS in cattle but there are few studies that indicate the optimal values of THI for buffalo. A value of THI < 72 is considered optimal, THI between 72 to 79 is considered as mild stress, 80–89 is considered as moderate stress, and ≥90 is considered as severe stress in buffalo [21]. When the maximum THI increases above 72, mild stress conditions in buffalo are reflected in a slight decline in milk yield [21].

Considering the effects of THI on the reproductive performance of buffalo, Dash et al. [22] classified the months of the year into two categories based on the THI values: a non-heat stress zone (NHSZ) and a heat stress zone (HSZ). The months from October to March were included in the NHSZ with the THI ranging from 56.71 to 73.21, and the months from April to September were considered in the HSZ with the THI ranging from 75.39 to 81.60. Within the HSZ, a critical heat stress zone (CHSZ) was also characterized. The CHSZ corresponded to the months of May and June and the THI ranged from 80.27 to 81.60. Choudhary and Sirohi [21], based on an analysis of their data set, reported the maximum, minimum, and average THI threshold levels for buffalo: the cut-off values were 74.37, 61.73, and 68.15, respectively. The threshold level indicates the critical level of THI up to which the animal can tolerate HS and after which there is a significant productivity decline. Thus, in buffalo, although a decrease in milk production was perceptible after the maximum THI exceeded 72, the decrease was significant only when the THI exceeded 74.

Under HS conditions, buffalos modify various physiological parameters, such as their rectal temperature (RT), respiratory rate (RR), heart rate (HR), skin temperature (ST), and body temperature (BT) [23]; these are considered the main physiological parameters that can be quantified to evaluate the presence of stressful conditions, such as HS, in animals [9]. It has been observed that buffalo did not modify their BT, HR, and RR compared to cattle in barns when the air temperature ranged from 30–33 °C with low (33–38%) and high (82–88%) humidity [19]. However, if buffalos were in the scorching sun for a long time, the BT, HR, RR, and general discomfort of buffalo increased more rapidly than those of cows [19]. Gudev et al. [24] reported that the exposure of lactating buffalo to direct solar radiation (THI = 77.83) causes a significant increase in their RT and RR, showing that the heat load is greater than the body's heat dissipation capacity. The same THI value did not induce significant changes in RT when buffalos were placed in the shade; although, maintenance of RT within the thermoneutral zone was achieved at the expense of a greater RR. A study carried out in Egypt [25] on buffalos and cows reported that under the same rearing conditions (the air temperature was 30–33 °C), the mean BT of both species was the same (38.8 °C), while

the mean RR was widely different (27.19 for the buffalo and the 46.69 for cows for respiratory acts for minute). Those authors reported that between 5 and 7 years of age, the RT significantly decreases by 0.90 °C in buffalo and 0.25 °C in cows and remains constant thereafter. The respiration rate also follows the same pattern as BT, with it tending to decrease with advancing age.

The symptoms of the buffalo at different THI ranges were reported by Dash et al. [22]: with THI values below 72, there was no stress condition and the buffalos were in an optimal condition for reproduction and production; for the THI range between 72–78, there was a mild stress condition and there was an increase in RT and RR; for the THI range between 79–88, there was a moderate stress condition, RR and water intake were significantly increased, dry matter intake was decreased, and the ratio of forage to concentrate intake was decreased; and for the THI range between 89–98, there was a severe stress condition. In this case, excessive panting and restlessness were observed. Rumination and urination were lowered along with a negative impact on reproductive performance in the buffalo; a value of THI above 98 is an extreme stress condition and buffalo may also die.

From the studies examined, due to their peculiar physical characteristics, buffalo suffers from HS, especially in tropical areas. The threshold levels of the THI are higher than those reported for cows. Once these levels are exceeded, they react by modifying some clinical, reproductive, and production parameters.

2. Mitigation Strategies

A reduction in the deleterious effects of HS is possible through acting in different ways. One strategy is the genetic approach by selecting subjects that are more tolerant to HS. For example, the Mediterranean Italian breed seems to be less tolerant than the Egyptian breed and the crossbreed (Nili-ravi × Murrah) [26][27]. Other approaches are feeding, nutritional, and cooling strategies (**Figure 1**).

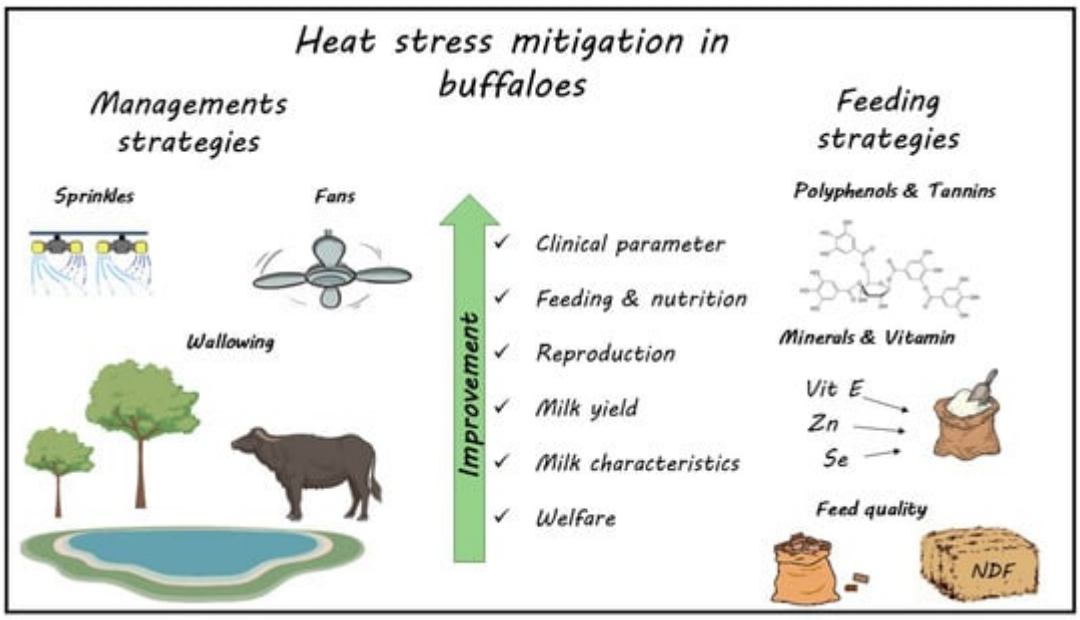

Figure 1. Heat stress mitigation strategies in buffalo.

2.1. Feeding and Nutritional Strategies

Different nutritional strategies have been implemented (**Table 1**). Some of them [28][29][30] supplemented the diet with minerals and/or vitamins that act like antioxidants, i.e., chemical substances that counteract the production of free radicals formed during exposure to HS [31]. Wafa et al. [32] supplemented the diet with *Moringa oleifera* leaves (MOL) which contained tannins, glycosides, anthocyanin, thiocarbamates, and polyphenols that can protect against oxidative stress. Moreover, Lakhani et al. [33] and Talukdar et al. [34] tested different diets in which there were different levels of fiber, protein, and energy with the aim to test diets with different metabolic heat production potential in the rumen.

Table 1. Effect of different nutritional and feeding treatment on several parameters.

References	Breed	Treatment	RT	ST	RR	HR	DMI	MY	MQ
[29]	Med. Italian	Zn + Se					n.e.	↑*	n.e.
[30]	Surti	Vit B_3	↓*	↓*	↓*		n.e.	↑*	↑*
[32]	Egyptian	*Moringa oleifera* leaves	↓*	↓*	↓*	↓*			
[33]	Murrah	34.5% NDF and 7% MP	n.e.		↓*	n.e.	n.e.	n.e.	↑*
		37% NDF and 7% MP	n.e.		n.e.	↓*	n.e.	n.e.	↑*
		30% NDF and 8% MP	↓*		↓*	↓*	n.e.	n.e.	↑*
		34.5% NDF and 8% MP	↓*		↓*	↓*	n.e.	n.e.	↑*
		37% NDF and 8% MP	↓*		↓*	↓*	n.e.	n.e.	↑*
[34]	Murrah	+15% ME	n.e.		n.e.	n.e.			
		−15% ME	↑*		↑*	n.e.			

n.e. no effect; ↑ increase; ↓ decrease; * $p < 0.05$. RT rectal temperature, ST skin temperature, RR respiration rate, HR heart rate, DMI dry matter intake, MY milk yield, MQ milk quality, NDF neutral detergent fiber, MP metabolizable protein, and ME metabolizable energy.

Evangelista et al. [29] and Chaudhary et al. [30] found that treatment did not affect dry matter intake but positively influenced MY with an increase in 3.5% and 10.9% L/day, respectively. Evangelista et al. [29] did not find any significant differences in milk fat, protein, and lactose content. In contrast, Chaudhary et al. [30] observed an increase in fat, protein, and lactose percentage. Moreover, Evangelista et al. [29] examined milk coagulation traits and observed the positive effects of treatment on milk coagulation properties. Chaudhary et al. [30] and Wafa et al. [32] found positive effects for treatment on physiological parameters showing a significant reduction in RT, ST, and RR (**Table 1**). Wafa et al. [32] also examined the effect of MOL on semen quality and reported a significant increase in the motility and viability of the semen and a decrease in the incidence of abnormality and damaged acrosome.

Other authors [28] fed the buffalo with supplementation of vit E + sodium selenite and observed that the pregnancy rate improved from 62.5% to 75.0% during the summer season.

Kumar et al. [35] administered an intra-muscular injection of vit E + Se and reported an increase of more than 3 kg/d in the DMI and an increase in MY of 1 kg/d and milk fat percentage (+0.5 percentage points) in the treated group compared to the control group. In addition, there was a decrease in RT (−1 °C), RR (−3 breaths/min), and HR (−5 beats/min) in the treated group compared to the control group.

These findings show and validate the importance of antioxidant supplementation during HS.

Lakhani et al. [33] found no effects of the different levels of fiber and proteins on the DMI and MY, but there was an improvement in milk quality. All of the diets increased the fat percentage, with the best value of 6.81 ± 0.05 for the diet containing 37% NDF and 8% metabolizable protein (MP) compared to the control diet containing 30% NDF and 7% MP. In addition, those authors observed a reduction in HR from 50 beats/min in the control diet to 46 beats/min in all the diets containing 8% MP. The treatment was also able to decrease the RT (about −0.2 °C) and RR (about −3 breaths/min). The best diet that improved the physiological parameters was that containing 34.5% NDF and 8% MP.

In addition, Talukdar et al. [34] investigated the effects of two different diets on physiological parameters and observed that increasing the metabolizable energy by 15% did not have any effect on RT, ST, and RR. In contrast, the reduction by 15% of metabolizable energy resulted in an increase in RT and ST; this was probably due to greater metabolic heat production derived from the increase in fiber content in the diet.

2.2. Cooling Strategies

A strategy to prevent HS in dairy buffalo is an intervention in their housing system and the adoption of cooling systems (**Figure 1**, **Table 2**). Several authors have ascertained the positive effect of cooling on the various parameters indicating the well-being of the animals in both temperate and tropical climates [36][37][38][39][40] and in both female and male buffalo [41]. These systems may affect several aspects of buffalo breeding including reproduction. Neglia et al. [37] observed in multiparous lactating Mediterranean Italian buffalo that a greater availability of space and the presence of a swimming pool improved the conception rate within 120 days post-partum (53.7% vs. 39.9% in the group with a pool and without a pool, respectively). Furthermore, the animals with a swimming pool showed a lower calving–conception interval than the group without a swimming pool, especially in the hottest months (April–August).

Table 2. The effect of different cooling strategies on several parameters.

References	Breed	Treatment Type	FB	CP	SI	MY	MC	RC	DMI	WI
[36]	Mediterranean Italian	Free stall open-sided barn with an outdoor lot and a concrete pool	n.e.		↑**	↑*	n.e.	n.e.		
[39]	Nili-Ravi	Shade with a fan				↑*	↑*		↑*	↓*

References	Breed	Treatment Type	FB	CP	SI	MY	MC	RC	DMI	WI
[14]	Murrah	Shade, a fan, and sprinklers				↑*	↑*		↑*	↓*
		Misting		↓*		↑*				
		Wallowing		↓*		n.e.				
[42]	Murrah	Silvopastoral system	↑*	↓*						
[38]	Murrah	Wallowing		↓*		↑*			↑*	
[40]	Nili-Ravi	Shade with a fan [36][43][44][45]		↓*		n.e.	↑*		↑*	↓*
		Shade, a fan, and sprinklers		↓* [46]		↑* [41]	↑*		↑*	↓*

n.e. = no effect; ↑ = increase; ↓ = decrease; * $p < 0.05$; ** $p < 0.01$. FB feeding behavior, CP clinical parameters, MY milk yield, SI social interaction, RC reproductive characteristics, SQ semen quality, DMI dry matter intake, and WI water intake.

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a day can improve the quality of sperm. They tested the effect of multiple showers on sperm quality in males under controlled climatic chamber conditions (the THI medium was 72.66 ± 2.30 in the morning and 81.66 ± 2.51 in the afternoon). The treated group was given four showers per day and the control group was given only one shower. The treated group was affected positively for the ejaculated volume (mL), live sperm (%), and sperm concentration (million cells/mL). They also found a higher percentage of normal fraction in the treated group compared to the control.

The presence of a pool improves both the quantity and quality of the milk and improves the heat dissipation capacity of the animals. It has been seen that swimming seems to be a more beneficial method than showers [38]. The research carried out by Aggarwal and Singh [38] on the comparison of two types of cooling systems for buffalo at the beginning of lactation clearly indicated that the group with the swimming pool presented greater feed intake and MY during the hot–dry and hot–humid seasons (the THIs were 80.3 and 83.6 during the hot–dry and hot–humid seasons, respectively). Moreover, the RT and RR were lower in the wallowing group especially during the evening time.

De Rosa et al. [36] on multiparous buffalo (3.20 ± 0.25 number of lactation) reported that the pool had a positive effect on MY (an increase of 1.8 kg/head in the hot period, i.e., the month of July with an average daily temperature of 24 °C). The characteristics of the milk were not influenced (fat and protein content), but the milk from pool group showed a greater tendency for higher somatic cells. The behavior of the buffalo was also affected by the presence of a pool. The pool group showed greater socialization activity between individuals (sniffing and nuzzling) and more agonistic interactions compared with the group without a pool. Therefore, the animals without the possibility of having an effective cooling system were affected by HS which determined the reduction in potential milk production and reduced their overall well-being. The authors, in fact, argued that the effective heat dissipation through wallowing by the buffalo with a pool may have contributed to sustaining the buffalo MY.

Other researchers [14] compared the wallowing system with misting over three months period (May, June, and July and the average THI of these three months was 79.88, 80.57, and 85.36, respectively). Milk production decreased in both experimental groups, but the decrease was more evident in the misting group. Rectal temperatures and RR

decreased in both groups compared with the control group but decreased more in the wallowing group in July. The heart rate increased in the wallowing group. Those authors concluded the wallowing and misting were equally effective in preventing a decline in MY during May and June (the hot–dry period); however, wallowing was highly effective during July (the hot–humid period) in maintaining MY.

Recently two similar studies, on Nili-Ravi buffalo, were carried out to test three different cooling methods; the first study was carried out during a hot–humid summer [39] and the second study was carried out during a hot–dry summer of a subtropical region [40]. The three systems adopted were: shade (S), shade with fans (SF), and shade, fans, and sprinklers (SFS). The results were similar in both climates. Greater MY was observed in the SF and SFS than in the control in both studies. The milk quality was better (fat, protein, and solid no fat) for the SF and SFS than the control, while no difference was found for the lactose content for the three groups in the humid climate [39]; instead, in the dry climate [40], the lactose content was also higher in the SF and SFS than in the S group. Under a hot–dry climate [40], a greater DMI in both of the treated groups (+17.34% and +5.40% for SFS and SF, respectively compared to the S group) was observed. Water intake was greater in the control group than in the two treated groups. The same results were also obtained during a hot–dry summer season [40]. In the hot–humid climate, several parameters were reduced such as the RR (reduced by 24.70 and 42.47% breaths per minute in the SF and SFS groups, respectively, compared to the control), the pulse rate (reduced by 10.36% and 23.32% beats per minute the for SF and SFS, respectively, compared to the control), and the ST (reduced from 0.87 in the SF and 2.09 °C in the SFS compared to the control).

Ahmad et al. [40] also investigated the effects of three cooling systems (S, SF, and SFS) on the feeding behavior of buffalo during a hot–dry summer climate. They found changes in eating activities. The eating time (min/24 h) was influenced by the treatments increasing from S to SFS (246.33, 280.33, and 309.50 min/days for S, SF, and SFS, respectively). The total time spent in drinking (min/24 h) the S, SF, and SFS was 24.67, 22.50 and 19.50, respectively. In addition, finally, the time spent in rumination was higher in the SFS (399.00 min/24 h) followed by the SF (385.17 min/24 h) and S (360.83 min/24 h). Furthermore, they observed that the total time spent in lying showed a greater value in the SFS than in the S. The time spent in locomotion (min/24 h) was 92.50, 76.33, and 68.67 for S, SF and SFS, respectively. All of these changes indicate greater well-being of the buffalo treated with SFS compared to the buffalo treated with SF and those treated with only S. Those authors concluded that the best cooling protocol to be used is the combination between shade, a fan, and sprinklers.

Recently, silvo-pastoral systems have been proposed as an alternative method to conventional breeding [42][47]. Athaide et al. [42] comparing two herds of buffalo reared one under a silvo-pastoral system (SPS) and the other one under a conventional system (CVS, pens with no shade) and found that subjects in the CVS had higher RT, ST, and HR. Galloso-Hernández et al. [47] carried out a study on heifers under two rearing systems: SPS and CVS. They reported that in the SPS, the ambient temperature was 2 °C lower than in the CVS (33.01 vs. 31.00 °C), and the feeding behavior (time spent eating, ruminating, and drinking water) was greater in the SPS under intense HS conditions (>75 THI). Under the CVS, the animals spent more time wallowing compared with the SPS. Galloso-

Hernández et al. [47] concluded that the time animals spent for thermoregulation was greater in conventional systems than in the SPS.

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