



Nutritional management of dairy animals for sustained production under heat stress scenario

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ABSTRACT

Dairy industry plays a significant role in the agriculture sector for sustainable growth. Heat stress, on the other hand, has been proven to have a detrimental impact on dairy output in terms of growth, reproductive performance and milk production in dairy animals, especially in tropical areas. Heat stress is one of the most significant issues facing the dairy industry, as rising temperatures and humidity limit animal productivity throughout the summer, resulting in devastating economic repercussions. The purpose of this review is to gather knowledge on the effects of heat stress on dairy output and how to ameliorate them. The diversion of energy resources from the production to the adaptation pathway may be responsible for the loss in productive capacity of dairy animals, when they are exposed to heat stress. There are different approaches pertaining to relieving the adverse effects of heat stress on dairy production system. These approaches may be classified into three major categories viz. genetic, management and nutritional interventions. These approaches might help dairy animals to perform better by reducing the harmful impacts of heat stress. Appropriate shelter design, giving shade, employing sprinklers, installing cooling devices, and using fans and ventilation systems are among the management strategies. The nutritional interventions comprise ration balancing and providing essential micronutrients to improve the productive and reproductive performance. Some of the most widely used dietary measures to ensure optimal production are inclusion of protein or fat (prill fat), micronutrients antioxidants (vitamins and minerals) and some feed additives (Astaxanthin, betaine, melatonin, *Chlorophytum borivilianum*) in the diet. These antioxidants and feed additives can be used to attenuate the negative effects of environmental stress. Furthermore, providing adequate energy and antioxidants help to ensure optimum growth, milk production and reproduction efficiency during heat stress. This review provides an overview of the consequences of heat stress on dairy animals, emphasizes essential nutritional strategies for heat stress reduction in dairy animals, and evaluates the influence of various feed supplements on growth, productivity and physiology.

Keywords: Amelioration, Antioxidants, Heat stress, Sustained production

In emerging countries like India, livestock plays an important role in the agricultural sector for sustainable growth. As per National Accounts Statistics 2020, the share of livestock in the agriculture and allied sector in terms of Gross Value Added (GVA) is estimated to be growing steadily from 24.32% (2014-15) to 28.63% (2018-19). Livestock sector contributed 4.19% of total GVA in 2018-19. The compound annual growth rate of livestock GVA is 6.23%, which is much higher than that of agriculture and allied sectors (2.02%). The global demand for animal-based consumables is increasing because of population growth, necessitating the continued expansion of livestock. By 2050, the global demand for livestock products is predicted to have doubled (www.faostat.org). However, there is a decline in production performance and affecting the population of livestock due to changes in environmental variables (climate change) that inflict stress on animals. Summer temperature

have been rising globally because of climate change, and this trend is expected to continue in the near future. According to the IPCC (2007), the rise in global average surface temperature might range from 1.8°C to 4.0°C by 2100. In addition, India has recently witnessed higher average temperature and humidity, as well as longer bouts of extreme heat, which is affecting livestock population and production performance. While farm animals suffer from many stresses during their lifespan, but heat stress is the most prevalent and tough to manage (Joy *et al.* 2020). High ambient temperature, direct and indirect sun radiation and humidity are all environmental stressors that put animals under strain. Heat stress, which is defined as the sum of external forces acting on an animal to create an increase in body temperature and provoke physiological reaction, is one of the most important environmental variables affecting livestock production performance (Dikmen and Hansen 2009). Because of the related decline in milk production and huge economic losses, heat stress has become a major concern for dairy producers (Polsky and Keyserlingk 2017).

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The consequences of climate change are projected to have a significant influence on dairy cattle production. Heat stress has a significant impact on the health and biological functioning of dairy cows, resulting in decreased milk production and reproductive performance (Polsky and Keyserlingk 2017). It also has several negative consequences on reproductive tissues, including loss of germ cells, poor morphology and quality of sperm and an irregular structure of DNA and chromatin (Erenpreiss *et al.* 2006). According to Sordillo and Aitken (2009), oxidative stress is the primary cause of immunological and inflammatory dysfunction.

Heat stress, caused by high ambient temperature and humidity, has a negative impact on farm animal's growth, output, and reproduction (Pandey *et al.* 2014). Thermal stress reduces animal feed intake, lowering productivity in terms of milk yield and quality (Singh and Upadhyay 2009), and reproductive performance, making it a considerable financial burden (St-Pierre *et al.* 2003). The livestock has the capacity and buffers to bear negative impacts of climate change and variability which makes it imperative to address the concerns for nutritional security in developing nations. Further, there is need of some of key factors to ameliorate heat stress and to maintain growth and production of cattle and buffaloes under changing climate. Most commonly, physical modification of the environment, breed selection for heat resistance and nutritional management are main strategies. This review provides an overview of the consequences of heat stress on dairy animals, emphasizes essential nutritional strategies for heat stress reduction in dairy animals, and evaluates the influence of various feed supplements on growth, productivity, and physiology during heat stress.

Heat stress effects on dairy animals

Feed intake: Animals exposed to high ambient temperature increase their effort to eliminate body heat, resulting in increased respiration rate, body temperature and water intake while decreasing feed intake (Marai *et al.* 2007). The quantity and quality of feed available to livestock grazing in a hot semiarid climate vary dramatically throughout the year (Martin *et al.* 2008). In addition to decreased feed consumption, substantially feed conversion efficiency is decreased. When an animal is exposed to a high ambient temperature, the peripheral thermal receptors send inhibitory nerve impulses to the hunger center in the hypothalamus, resulting in a decrease in feed intake (Marai *et al.* 2007). During heat stress, dry matter intake is decreased by 9 – 13% depending upon the intensity and duration of exposure of dairy animals. The decrease in feed consumption could be attributed to the animal's adaptive mechanism. Panting, which inhibits cud-chewing, slows feed digestion, and limits the quantity of water and buffers from saliva reaching the rumen, is primarily responsible for the lowered feed intake.

Growth is governed both genetically and environmentally in terms of the increase in living body mass

or cell multiplication. High ambient temperature is a factor affecting growth and average daily weight gain in animals. A decline in anabolic activity and the increased tissue catabolism during higher temperature can be attributed to decreased growth (Marai *et al.* 2007). Further, the rise in tissue catabolism can be explained by the increase in catecholamines and glucocorticoids following the exposure of animals to heat stress. During early lactation and high-yielding cows are more severely affected. Standing in the shade to keep cool also reduces grazing time and feed intake in these animals. Reduced feed intake during thermal stress may account for up to 50% of the loss in milk output, while the other 50% may be influenced by heat-related lactogenic hormone variations (Johnson 1987). Reduced feed intake during heat stress accounts for 60% of decreased milk yield (Almoosavi *et al.* 2021).

Milk yield

The impact of high temperature on milk production is likely to be the most detrimental to any animal production system that pushes animals to limit feed intake, resulting in lower milk yield (Dunn *et al.* 2014). The decrease in milk production is mostly related to the reduced feed intake. If the water becomes limited, milk production will decline even more. Animals redirect bodily water from milk production to cool themselves by sweating and panting. Heat stress decreases milk production from 10 to > 20% in Indian conditions (Singh and Upadhyay 2009). In the heat stress cows, milk output dropped by 21%, showing that lower feed intake during late gestation accounted for 60% of the overall reduced milk production (Almoosavi *et al.* 2021). In addition to high temperature, humidity is an important factor that causes more stress and decreases production performance. Heat stress affected milk production, with a negative association between milk yield and temperature-humidity index (THI). Igono *et al.* (1992) studied the effect of ambient temperature on milk production of Holstein cow and reported an inverse relationship among them. THI was used by Ravagnolo and Misztal (2000) to investigate the effect of heat stress on milk production. They found that when THI surpassed 72, milk yield decreased by 0.2 kg per unit rise in THI. Bouraoui *et al.* (2002) found that increase in THI is inversely related to milk production, with an increase in THI from 68 to 78, there is a decrease in milk production by 21% and DMI by 9.6%. Malmkvist *et al.* (2009) reported higher catecholamine levels in blood due to heat stress which inhibited oxytocin release and ultimately controls milk ejection. Heat stress not only reduces milk output in dairy animals, but also has a negative impact on milk quality. Aggarwal and Singh (2007) reported a change in the composition of milk in buffaloes during thermal stress conditions. The fall in fat percentages could be associated with a decrease in the consumption of forage (17%), which could result in insufficient fiber content in the diet for appropriate rumen function. According to Rhoads *et al.* (2009), a drop in DMI could account for 30 - 50% of a drop in milk production in heat-stressed cows.

Rejeb *et al.* (2012) reported a substantial ($P<0.01$) drop in milk fat content owing to heat stress, ranging from 3.79 percent in the spring (THI=65.62) to 3.65% in the summer (THI=83.91). Drop in milk was more in crossbred (KF) cows compared to indigenous (Sahiwal) cows (Vaidya and Singh 2019). Kumar *et al.* (2020) also reviewed that heat stress adversely affects production performance of high yielding dairy cows in terms of quantity as well as quality of milk.

Reproduction

Fertility rate of dairy animals are compromised during summer compared to the winter season. Reproductive processes in ruminant species have been impacted by heat exposure (Naqvi *et al.* 2004) and the inhibitory effects of stress on reproduction are mainly mediated by glucocorticoids (Kornmatitsuk *et al.* 2008). Substantially there is a decrease in the level of one of the primary reproductive hormones (estradiol) during heat stress conditions. The effect of high ambient temperature on the hypothalamic-pituitary-ovarian axis plays an important role in regulating hormonal secretion, behavior, follicular development and embryonic growth and survival in the uterus of animals. These changes alter key components of livestock breeding, such as the duration of the estrus, fertility and conception rate (Penev *et al.* 2021). Nutrition is widely known to regulate reproductive endocrine functioning in many species. Nutrient insufficiency caused by low feed intake after heat exposure may affect the reproductive processes, influencing estrus behavior and ovulation rate. By interfering with reproductive tract physiology, heat stress affects bovine fertility and reproductive efficiency, resulting in decreased oocyte production or low semen quality, as well as lower embryo development and survival (Krishnan *et al.* 2017). During heat stress, there is a decrease in immunity resulting in the occurrence of disease, which may also compromise reproductive efficiency in dairy animals. Changes in ambient temperature have an impact on animal fertility and reproductive performance by interfering with reproductive tract activities, disturbing hormonal balance, reducing oocyte formation, and consequently limiting embryo growth and survival (Collier *et al.* 2006). Heat stress

has been shown to have a considerable impact on the ability of spermatozoa to penetrate oocytes, as well as subsequent embryo development and quality (Erenpreiss *et al.* 2006). Roth *et al.* (2001) reported that heat stress decreases the follicle size of the first and the second follicular waves and suggests that bovine exposure during thermal stress modifies follicular dynamics and diminishes follicular prevalence during the next period. Heat stress is the leading cause of bovine infertility and reproductive inefficiency, both of which contribute to significant economic losses.

Strategies to ameliorate heat stress for sustained production

Physical microclimate alteration, heat-resistant breed selection and nutritional management are the three most important factors in maintaining productivity during heat stress. Summer stress can be ameliorated through a variety of methods, such as management techniques (shading / boosting ventilation by utilizing fans) or increasing heat loss from the body i.e. cooling systems (Singh *et al.* 2014); open pen-ridge ventilated thatched roof housing (Mandal *et al.* 2021) or additional water troughs, dietary modification (low fiber and low protein), increasing nutrient concentration in the feed to compensate for lower intake, altering the rate of starch fermentation or increasing energy-dense material for enhancement of energy density of ration (Somagond *et al.* 2019), dietary vitamin E, Zn, CrPic, betaine and astaxanthin as an antioxidant to reduce oxidative stress caused by heat exposure.

Nutritional strategies

Nutritional modifications are primarily used to reduce the internal heat load on animals. A range of nutritional strategies can be used to reduce heat stress in dairy cattle. During the summer, there is a decline in nutritional feed intake, which is responsible for decreased production performance. Effective practical approaches such as frequent feeding, use of higher quality forage, appropriate nutritional balance and better nutrient density in the ration are needed in these situations (Sejian *et al.* 2012). Feed fresh, palatable and high-quality forages to animals and feed ingredients should have high digestibility to lower the heat increment during digestion (Chase 2006). The amount of

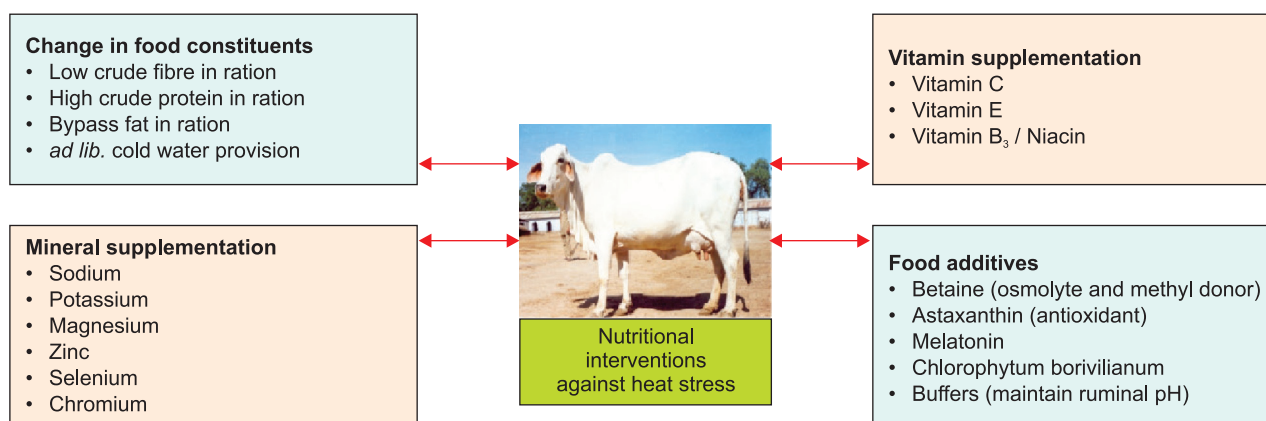


Fig. 1. Nutritional strategies to ameliorate heat stress in animal.

metabolic heat production is affected by the quality as well as the particle size of feeds and fodder. Simple feed technologies like incorporation of good-quality green fodder, increment of nutrient density by substituting low roughage quality with concentrate and feeding of appropriately chopped dry fodder and hydration of dry straws during hot dry period reduce the internal heat load on the animal body. Some of the feed additives like antioxidants and minerals can also be supplemented to minimize the impact of climatic extremes. To counteract the negative effects of heat stress, dietary supplements such as vitamins, trace elements, and minerals are used exclusively. Khorsandi *et al.* (2016) reported that feeding of heat-stressed transition cows with ruminal bolus increased milk production, suggesting that it ameliorates the adverse effect of stress. Supplementation of feed additives i.e., antistress vitamins like vitamin C, A, E, and Niacin, buffers (Chase 2006), high-density diet (Somagond *et al.* 2019), herbal supplements like betaine (Deshpande *et al.* 2020), astaxanthin (Somagond *et al.* 2020) and *Chlorophytum borivilianum* (Devi *et al.* 2021a) are also used to ameliorate the effect of summer stress and for sustained production. The nutritional strategies of high-producing dairy cows require additional attention to fulfill their total demand for milk output, health and reproductive potential (Roche *et al.* 2013). Some of the nutritional strategies tested under farm and field conditions for the amelioration of heat stress are explained in Fig. 1.

Fiber level in the ration

Fiber and its digestibility are the first nutrients that interact with hot weather conditions. It is generally understood that fiber can influence voluntary dry matter intake (DMI), chewing, and ruminating activity (Conte *et al.* 2018). Level of crude fiber (CF) in ration plays important role in heat stress amelioration. Also, sufficient fiber content in the ration is must for normal rumen conditions as well as high quality forage maintains feed intake normal. NRC (2001) provided recommendation of minimum 25% dietary neutral detergent fiber (NDF), out of that 75% of NDF from roughages. But digestion as well as metabolism of fiber portion of ration generates more heat than concentrates. Feeding of the low crude fiber (12.50% on dry matter basis) containing ration to lactating cows resulted a significant decrease ($P < 0.01$) of rectal temperature, pulse rate and respiration rate. In the hot humid tropical regions, fiber levels during ration formulations of ruminants should be considered. West *et al.* (2003) reported that cows fed a low-fiber diet (NDF = 30% of DM) during hot weather had higher daily milk output, lower body temperature and respiration rate than cows fed high-fiber diet (NDF = 42 percent of DM). Further it was evidenced that by decreasing NDF level in diet helped in lowering the respiration rate and rectal temperature and increase in DMI and milk production of dairy cows (Miron *et al.* 2008). Similarly, Adin *et al.* (2009) reported that by reducing dietary NDF from 18 to 12% on DM basis was responsible for a

significant decrease in rectal temperature. Conte *et al.* (2018) reviewed that low metabolic heat increment diets can aid to enhance feed intake and performance under heat stress circumstances. Fiber intake has a significant impact on the heat generation, and its importance must be taken into account when developing an efficient nutritional and environmental management strategy.

Protein level in the ration

Heat stressed dairy animals often undergo negative energy balance (NEBAL). Heat stress leads to reduced feed intake, therefore dietary crude protein (CP) levels in ration should be increased during heat stress conditions. Rumen undegradable protein (RUDP) (bypass protein); part of the dietary CP; must be increased in diet, because the net passage rate of microbial protein from the rumen drops due to reduced dry matter (DM) intake. There are also some variations within the literature as benefits vs negative concerns of improved protein and changed protein solubilities (Huber *et al.* 1994). Addition of more dietary CP, specifically RUDP, is not useful during heat stress conditions (Arieli *et al.* 2004). Low rumen motility and declined passage rate due to highly degradable protein diets may be a possible reason of deleterious effects during heat stress (Linn 1997). Supplementation of methionine boosts milk production and antioxidant capacity and lowers lymphocyte apoptosis (Han 2009). Conte *et al.* (2018) reported lower DMI during heat stress, therefore it is important to increase the amount of protein in the diet. Further, it is vital to feed rumen undegradable proteins or enhance protein quality by increasing the quantity of key amino acids (especially methionine and lysine). Neutral detergent fibers (NDF) @ 34.5% and metabolizable protein (MP) @ 8.4% in ration of lactating Murrah buffaloes have beneficial effect on heat stress amelioration (Lakhani *et al.* 2021).

Fat supplementation

The method to cut down the production of metabolic heat was commonly adopted with higher dietary fat in the ration because fat has a lower heat increment (up to 50%) than forages, extra fat should be supplemented and the fiber component of the diet should be reduced. When compared to other nutrients such as fiber and carbohydrate, the inclusion of fat in diets may reduce the heat load due to its high energy density and reduced metabolic heat. Melo *et al.* (2016) found that cows supplementing with palm oil had lowered rectal temperature and respiratory frequency, increased milk output and feed efficiency. Feeding of calcium salt of palm oil fatty acids considerably ($P < 0.05$) maintained body condition compared to the control group of Murrah buffaloes (Mane *et al.* 2017). Feeding 4% Ca salts of fatty acids to Murrah buffaloes lead to an improvement of 12.4% in their milk yield (Shelke *et al.* 2012).

Bypass fat supplementation to early lactating animals increased the energy density in ration and prevents the acute

negative energy balance during heat stress (Tyagi *et al.* 2009). One such bypass fat is 'Prill Fat'. Prilled fat is non-hydrogenated vegetable oil, containing more than 85% palmitic acid that bypasses rumen fermentation (Singh *et al.* 2015). The product is hard fat with a melting point $>57^{\circ}\text{C}$, it resists lipolysis and biohydrogenation by rumen microbes but gets digested in the lower tract and can be added up to 9% of dry matter intake due to its inertness. Crystalline or prilled fat can be produced by liquefying and spraying the saturated fatty acids under pressure into the cooled atmosphere, thereby increasing the melting point of the fatty acids and not melting at ruminal temperature, thus resisting rumen hydrolysis and associating them with bacterial cells or feeding particles (Grummer 1988). Rumen-protected fat increases caloric density without reducing the content and digestion of dietary fibers (Schauff and Clark 1989). In Indian feeding conditions, 75gm of prilled fat can be supplemented to lactating cows (Gnanasekar *et al.* 2016). To overcome the effect of negative energy balance, Ca salt of fatty acid has been used by many researchers for supplementing to dairy animals (Yadav *et al.* 2015). Pre-partum (100g/day) and post-partum (150g/day) fat supplementation resulted in a substantial increase in the milk yield and content of milk fat and enhanced BCS and energy balance in Murrah buffaloes without altering protein, lactose, and SNF (Sharma *et al.* 2016). In lactating dairy animals, bypass fat (200g/day) supplementation increased milk production and feed efficiency. In early lactating cows' milk production was enhanced by 6.02 percent, when they are supplemented with 75 g of prill fat per day (Rajesh *et al.* 2014). Similar amounts of prill fat feeding to Murrah buffaloes resulted in an increase of 10 percent in milk yield in the organized herd and an increase of 17 percent under field conditions (Singh *et al.* 2015). Dietary fat (Prill fat) along with astaxanthin (antioxidant) was more effective in ameliorating adverse effect of summer stress (Somagond *et al.* 2019, 2020). It can be stated that bypass fat supplementation to the dairy animals very important to alleviate negative energy balance during heat stress without adversely affecting the dry matter intake and rumen fermentation. Bypass fat supplementation provides additional benefits due to increased milk yield, nutrient utilization efficiency, postpartum recovery from negative energy balance and production performance during adverse climatic conditions.

Water

Ad lib. water availability is the critical for dairy animals when exposed to environmental heat stress. Water is an important nutrient for ruminants to cope with heat stress since it aids in animal thermoregulation. Water helps in dissipating excess body heat via sweating and panting mechanisms. Intake of water has positive correlation with milk production and DMI (Dado and Allen 1994). Intake of water rises with the rise in environmental temperature because of more water loss via sweating and panting. Water intake rises by 50% as the THI ≤ 80 . *Ad lib* supply of clean

water within shed is essential for dairy animals. Ice cubes or blocks can be added in drinking water to lower drinking water temperature during heat stress conditions (Sastry and Tripathi 1988). Under thermally severe conditions, the water intake of Holstein cows producing 28 kg of milk per day might range from 121 to 135 L per day and further reported dietary betaine was helpful in improving the milk production (Hall *et al.* 2016). HF lactating cows exposed to constant 32.2°C temperature showed marked increase in body surface moisture evaporation with 28% increase in water intake whereas 33% decrease in faecal water content (McDowell *et al.* 1969). Providing water of 10°C temperature to lactating cows reduced body temperature (0.75°C) than water of 28°C temperature (0.46°C). Water of 10°C temperature also reduced respiration rate and kept body temperature lower. Chilled water was only about 32% effective in reducing body temperature. Water restriction, after feeding, especially 2 hours, in heat stressed ewes improved nitrogen balance and nutrient digestibility without adverse effects on immunity (Nejad *et al.* 2014).

Buffers

Buffer's supplementation can be helpful during heat stress conditions. If fiber content of ration is reduced as well as animals are selectively eating concentrates, then buffers prevent a low ruminal content pH and rumen acidosis problems. Also, increased sodium content in diets which fed during heat stress has shown to increase DMI with milk yield. Buffers like sodium bicarbonate, magnesium oxide and sodium sesquicarbonate maintain a normal ruminal environment and lowers the chances of ruminal acidosis commonly occurs during heat stress (Chase 2006).

Mineral supplementation

Minerals are inorganic components that are necessary for metabolic processes, development, synthesis of milk, reproduction and health. Animals must receive minerals through feed since they cannot produce them in their bodies. Feed and fodders are a poor source of minerals; they do not supply all of the mineral's necessary for normal physiology and functions of animals. As a result, animals' rations should include an appropriate amount of high-quality minerals combination. Minerals are now available in different forms (chelated, nano and organic minerals) and also trace minerals are now being attached to oligopeptides to make them more bioavailable. Minerals supplementation in hot climates must be seen not just as a simple means of covering the requirement of a specific nutrient, but also as a means of mitigating the influence of heat stress during the summer season (Calamari *et al.* 2011). Micronutrient (Vitamin A, E and Zinc) supplementation during the prepartum period can enhance dairy cow's health and as a result improved the well-being of their calves (Alhussien *et al.* 2021). Similarly, the inclusion of antioxidant micronutrients (Cu, Zn and vitamins A and E) in to the buffalo rations enhanced udder health by lowering

the incidence of mastitis and improved milk yield as well as milk fat and protein percentage (Singh *et al.* 2021a).

Sodium, potassium and magnesium

To maintain water balance, ion balance and acid-base status of heat-stressed animals, electrolyte minerals, sodium (Na) and potassium (K) are necessary. For a variety of crucial physiological processes, sodium together with chlorine and potassium in proper concentrations are necessary. Potassium (K⁺) is the main osmotic regulator of sweat gland production in dairy animals. Heat-stressed cows require more Na to replace losses because of an increase in the sweating rate. Sodium chloride (NaCl) is a low-price supplement containing high Na levels (39.3%), which may enhance the milk output of dairy animals during heat stress. It was also observed that excessive levels of sodium and potassium chloride salts affect the heat-stressed animals digestion, acid-base and mineral status of dairy animals. Specific nutritional requirements for milk production, such as Na and K, appear to be different under heat stress than under thermoneutral state. Dietary requirement for K during summer is increased from 1.4 to 1.6% of DM. Further, as sodium (Na⁺) and magnesium (Mg⁺) compete with K⁺ for intestinal absorption, their concentration in diet should be increased. When feeding greater quantity of potassium, provide magnesium at 0.35-0.4 percent DM to assist minimize metabolic disease. According to Sanchez *et al.* (1994), NaHCO₃, K₂CO₃, and KHCO₃ increase milk production and fat content in dairy cows by improving feed intake and performance during hot weather. The rumen is apparently buffered, and a higher ruminal pH is maintained by providing 0.85 to 1.0 percent dietary sodium bicarbonate and it also maintains acid-base homeostasis in thermally stressed cows.

Zinc

Zinc is an enzyme activator that regulates pH, immunological competence, and fundamental cellular processes (Takkar 2011). Zn is the only lymphocytic mitogen that occurs naturally, its deficiency leads to immune dysfunction (Haase and Rink 2009). To ensure continuous immunological function, a suitable daily supply of zinc is essential, as there is no specific zinc storage system in the body. The influence of Zn on the antioxidant defense system and reducing oxidative damage on cellular components has been proven by several researchers (Picco *et al.* 2004, Sheikh *et al.* 2017). Zinc supplementation during the study period aid in reducing heat stress and improving immunity in transition dairy cattle (Sheikh *et al.* 2017). Zn in the basal feed at concentrations of 80 ppm and 120 ppm enhanced the reproductive efficiency of Karan Fries dairy cattle (Patel *et al.* 2017). Further, dietary Zn supplementation also showed lower cortisol levels, better immunity and increased productivity in Karan Fries cows (Patel *et al.* 2018). These findings suggest that zinc supplementation during the summer season attenuates the adverse effects and augments immunity of dairy animals.

Selenium

Selenium contributes to the maintenance of healthy endogenous antioxidant levels in the body's tissues, it is a component of the glutathione peroxidase (GPX) enzyme, which eliminates free radicals in the cytoplasm and helps to protects tissues from oxidative stress. Supplementation of selenium and vitamin E to dairy animals improves the deleterious effects of heat stress on the consumption of feed, physiological responses, acid-base balance and oxidative stress (Chauhan *et al.* 2014). Thatcher (2006) found an increase in immune competence, better uterus health and pregnancy rate in cows given selenium-yeast in the summer months during the transition period. Similarly, Calamari *et al.* (2011) found that greater antioxidative defense and improvements in immunity in cows subjected to heat stress are supplemented with selenium in diet. Furthermore, selenium improves average daily feed intake and growth while maintaining oxidative balance in lambs that are subjected to heat stress (Chauhan *et al.* 2016).

Chromium

Chromium plays a vital role in energy metabolism. Chromium picolinate and chromium nicotinate are the most common supplementary sources. In comparison to organic chelated chromium, inorganic chromium sources are poorly absorbed. Chromium improves tissue sensitivity to insulin and glucose consumption and acts as a glucose tolerance factor. Insulin resistance is linked to a decreased capacity to regulate body temperature; therefore insulin-resistant animals are likely more affected during stress (Dunshea *et al.* 2008). Chromium helps insulin to work more efficiently on glucose, lipid and protein metabolism. According to Spears *et al.* (2012), heifers fed with a higher dosage of chromium showed improved insulin sensitivity, suggesting that chromium has an important role in glucose metabolism in ruminants and helps to ameliorate heat stress. Chromium picolinate supplemented @1.5mg/ kg DMI to buffaloes showed less expression of HSP70, 90, 110, and lower NEFA level as compared to control (NICRA report IVRI). Zhang *et al.* (2014) found that chromium supplemented HF dairy cows had higher HSP72 levels. Supplemental Cr boosts immune function by increasing serum immunoglobulin synthesis, antibody titer to antigens, or by lowering blood cortisol levels and regulating inflammatory responses (Spears and Weiss 2008). Furthermore, feeding chromium to heat-stressed dairy cows reduced weight loss, enhanced milk output, decreased plasma NEFA concentrations, and increased pregnancy rate (Mirzaei *et al.* 2011). Kumar *et al.* (2015a) reported that dietary inclusion of chromium during heat stress improved the immunity and growth performance of buffaloes. T and B cell proliferation, neutrophil phagocytic activity and immunoglobulin content all improved when heat-stressed buffalo calves were given inorganic chromium (Kumar *et al.* 2015b). Microenvironment modification along with chromium supplementation can be suggested as an important management practice during heat stress for Karan Fries

cattle as it helped in increasing DMI and ADG and decrease water intake of growing calves and reduces the levels of stress indicator (cortisol, catalase), and enhanced immunity (IgA and IgM) (Sunil *et al.* 2017). Chromium supplementation @ 0.50 mg/kg dry matter improved glucose metabolism and maintained vaginal temperature in heat stress dairy cows (dos Santos Ribeiro *et al.* 2019). Chromium supplementation to high-yielding dairy cows during the transition period improved feed intake and milk production during early lactation. Further, it also improved reproductive performance, cell-mediated and humoral-immune responses.

Vitamin supplementation

Vitamin A, C, E and zinc supplementation showed positive effects in ameliorating environmental heat stress (MacDowell 1989). Vitamin C and E possess antioxidant properties.

Vitamin C

Heat stress leads to reduction in plasma vitamin C levels in lactating cows (Padilla *et al.* 2006). Ascorbate as well as zinc functions as scavengers of reactive oxygen species (ROS) during cellular oxidative stress. Supplementation of vitamin C and electrolyte showed ameliorating effects of the environmental heat stress in buffaloes (Kumar *et al.* 2010). Supplementation of ascorbic acid @ 10g/animal/day tended to shift the changes in immune status, biochemical parameters, hormones and physiological responses towards normalcy during heat stress in pregnant buffalo heifers (Ganaie *et al.* 2012). Feeding of rumen protected capsule containing vitamin C, K₂SO₄, niacin and GABA ameliorated environmental heat stress and improved dairy cow production (Guo *et al.* 2017).

Vitamin E

Vitamin E provides protection to the biological membranes against ROS damage, and it plays a role as an inhibitor 'chain blocker' of lipid peroxidation reactions (Seyrek *et al.* 2004). Suitable concentrations of antioxidants like selenium (Se) and vitamin E, are crucial not only for the maintaining health and efficiency of ruminants but also for the quality of milk production (Castillo *et al.* 2013).

Niacin / Vitamin B₃

Among vitamins, niacin has been evaluated for its effect on mammalian vasodilation and its function in lipid metabolism. Niacin increases peripheral vasodilation which leads to increase in sweat gland activity in farm animals (Gille *et al.* 2008). Niacin helps to relieve heat stress by boosting evaporative heat dissipation from the body as well as decreasing the effects of heat at the cell level (Lundqvist *et al.* 2008). These are the adaptive mechanisms for dissipation of extra body heat via increased sweating due to peripheral vasodilation. This helps to prevent reduction in DMI and finally improving milk yield. Di Costanzo *et al.* (1997) reported that cows exposed to mild to severe

heat stress conditions; fed encapsulated niacin @ 12 and 24 g/cow/day showed reduced skin temperature and increased milk yield. However, Yuan *et al.* (2011) showed that supplementation of rumen protected niacin @ 12 g/cow/day improved lipid metabolism. Zimbelman *et al.* (2013) found that dairy cows provided supplementation of encapsulated niacin @ 12 g/cow/day during acute heat stress leads to a lower core body temperature, increased sweating and variable effect on milk production.

Feed additives

Betaine: Betaine (trimethylglycine) is a natural, non-toxic and stable amino acid derivative found in many plants and invertebrate species. Physiologically, betaine is an osmolyte and a methyl group donor (Lever and Slow 2010). Cronje (2005) has recommended that heat stress and its negative effects on the gut and gut integrity can be improved by dietary betaine supplementation. Dietary betaine supplementation lowers heat stress effects with improving DMI and growth rate in beef cattle (Cronje 2005; Loxton *et al.* 2007) and sheep (DiGiacomo 2011). Betaine decelerates ruminal fermentation of starch and decreases the heat increment due to feeding and helps ruminants to handle environmental heat stress. Dietary betaine supplementation @ 50 g/cow/day during hot humid season resulted in improved reproductive performance of Karan Fries cows (Raheja *et al.* 2018).

A significant decrease was observed in RT, RR, ST and an increase in DMI, ADG, TBARS, Catalase, GPx, and SOD activity with increased total protein level in Karan Fries heifers fed with dietary betaine during hot dry as well as hot humid conditions (Lakhani 2018). A decrease in rectal temperature and increase of feed intake, milk yield, GPx and SOD activity was observed in Karan Fries cows supplemented with dietary betaine during heat stress (Raheja 2017). In comparison with control group animals, betaine supplementation to lactating HF cows at high environmental temperature increased average daily milk production ($P<0.001$). Also, dietary betaine increased milk protein levels ($P<0.001$) and milk fat concentration ($P<0.001$) (Dunshea *et al.* 2019). During heat stress conditions ($THI\leq 72$), supplementation of betaine to lactating cows showed significantly higher ($P<0.05$) DMI, milk production and milk fat % as compared to control. Somatic cell count (SCC) was higher ($P<0.05$) in the control group as compared to treatment group. The levels of volatile fatty acids (VFA) in the treatment group were significantly higher ($P<0.05$) than that of the control group. The concentration of serum total antioxidant capacity (T-AOC) in treatment group was significantly improved ($P<0.05$). Also, MDA and SOD of treatment group were higher ($P<0.05$) compared to control group. Compared to control, treatment group had higher ($P<0.05$) serum glucose. Cows given dietary betaine showed decreased serum BHBA as well as NEFA levels (Shah *et al.* 2020). Dietary betaine supplementation proved as a potent growth promoter by lowering the levels of plasma cortisol and NEFA and enhancing the ADG, DMI

of buffalo heifers. Also, dietary betaine supplementation increased plasma growth hormone levels with reduced physiological responses (Deshpande *et al.* 2020). Feeding of betaine hydrochloride to lactating buffaloes under heat stress increased DMI and milk fat%, whereas physiological responses like RT, RR and PR were reduced ($P < 0.01$) as compared to control group (Shankhpal *et al.* 2019).

Astaxanthin

Astaxanthin is a xanthophyll carotenoid naturally found in many crustaceans and red yeasts. It possesses anti-inflammatory, antioxidant, photoprotective and hepatodetoxicant properties. Astaxanthin enhances immune response by reducing the DNA oxidative damage and inflammation in young healthy adult female human (Park *et al.* 2010) and improved acquired immunity by T cell proliferation and IgG production in male broiler chicks (Takimoto *et al.* 2007). Astaxanthin can be utilized as a key feed additive that helped animals to cope up with the negative effects of climate change and stress, as well as to boost their productivity (Table 1) under stressful conditions (Singh *et al.* 2021b). There was a reduction in levels of the pro-inflammatory cytokines, such as tumor necrosis factor- α and interleukin-6 in the astaxanthin supplemented group (Macedo *et al.* 2010). Priyadarshini and Aggarwal (2018) showed lower ($P \leq 0.05$) expression of IL-6 and TNF- α during summer in Murrah buffaloes supplemented with astaxanthin. Kumar and Singh (2019) reported a beneficial effect of astaxanthin for reducing levels of pro-inflammatory cytokines in H_2O_2 -stimulated U937 mononuclear cells by hindering the ROS-induced production of NF- κB transcription factor. Katsoulos *et al.* (2009) reported that supplementation of Clinoptilolite (Astaxanthin 2.5% w/v) along with feed to dairy goats

showed improved milk fat percentage, besides reduced SCC suggesting improved milk quality. It was also suggested that 2% (w/v) Clinoptilolite supplementation for 120 days significantly improved milk yield in dairy cows (Alic 2014). Dietary inclusion of Astaxanthin @ 0.25 mg per kg BW per day per animal has heat stress ameliorative action in KF heifers (Kumar *et al.* 2019). Astaxanthin treatment reduced the negative effects of heat stress and increased the growth rate of heifers, allowing them to reach puberty earlier by conserving energy that would otherwise be needed to dissipate heat stress (Kumar and Singh 2020). HSP70 expression in periparturient buffaloes was reduced after dietary inclusion of astaxanthin, implying that supplementation reduces the negative effects of thermal stress by boosting immunity under stressful situations (Priyadarshini and Aggarwal 2018). Somgaond *et al.* (2019) reported supplementation of astaxanthin along with dietary fat (Prill fat) was more effective in reducing negative effects of heat stress and improving production performance of lactating buffaloes under heat stress conditions.

Melatonin

Melatonin quenches oxidants, particularly nitric oxides, because of its antioxidant characteristics, acts synergistically with antioxidants like glutathione peroxidase, superoxide dismutase, vitamin E and selenium (Tan *et al.* 1993, Gitto *et al.* 2001, Mauriz *et al.* 2013). Melatonin is a powerful antioxidant and free radical scavenger that regulates seasonal reproduction in photoperiodic species. Exogenous melatonin that is released continuously protects against oxidative stress, improves TAC concentrations, further it has shown the capacity to neutralize harmful Reactive Oxygen Species (ROS), lower lipid peroxide concentrations, minimize DNA damage and increased germ cell viability (Wang *et al.* 2013).

Table 1. Effect of astaxanthin supplementation on bovines during heat stress conditions

Animal	Dosage	Effects of astaxanthin	Reference
Karan Fries bulls	0.25mg/kg BW/day/animal	Improved semen quality during summer season	Soren <i>et al.</i> (2017) Soren and Singh (2018)
Murrah buffaloes (pre and postpartum)	0.25mg/kg BW/day/animal	Inhibited inflammatory mediators by blocking NF- κB activation. Improved milk production by 7 % during summer season	Priyadarshini and Aggarwal (2018)
Karan Fries and Tharparkar heifers	0.25mg/kg BW/day/animal	Weight gain and early attainment of puberty during summer season. Improvement in antioxidant status, immunity, growth rate and decreased the apoptosis rate	Kumar and Singh (2020)
Lactating Murrah Buffaloes	0.25mg/kg BW/day/animal	Increased milk yield by 5.39% by astaxanthin supplementation alone and 15.6% by a combination of astaxanthin + prill fat Reduced physiological responses and stress indicators levels during summer stress	Somgaond <i>et al.</i> (2019) and Somgaond <i>et al.</i> (2020)

Source: Singh *et al.* 2021b.

Melatonin acts by increasing the strength of antioxidants and lowering oxidative stress that would help in maintaining the reproductive efficiency of buffaloes during summer (Kumar *et al.* 2015a). Melatonin supplementation @ 18mg/50 kg body weight at 3-week interval has shown ameliorative effects i.e., lower expression of HSP 70 and 90 during summer, reduced rectal temperature and altered antioxidant enzyme profile in buffaloes (Kumar *et al.* 2015a). The melatonin supplementation @ 18mg/50 kg body weight at 3-week interval has shown ameliorative effect during summer as evident by reduced rectal temperature and altered antioxidant enzyme profile and enhanced the quality of buffalo bulls' sperm during the non-breeding season under tropical conditions (Ramadan *et al.* 2019). The behavioral symptoms of estrus in heat stressed post-partum anestrous buffaloes were quite high in melatonin implanted buffaloes.

Chlorophytum borivilianum

Chlorophytum borivilianum (CB) also known as 'Safed Musli,' is a well-known herb for boosting body immunity (Thakur *et al.* 2007). CB roots are rich in carbohydrates, protein, fiber, saponins and alkaloids (Singh *et al.* 2012). This herb has a wide range of therapeutic properties, and it acts as a total rejuvenator, antioxidant and immune modulator. This herb's antioxidant efficacy is attributed to its strong nitric oxide, superoxide and free radical scavenging activities (Visavadiya *et al.* 2010, Ahmad *et al.* 2014). It contains cytotoxic steroidal glycoside saponin chloromaloside-A and spirostanolpenta glycosides embracing beta-Dapiofuranose that are responsible for anticancer property (Qiu *et al.* 2000). CB root powder also improved antioxidant enzymes and vitamin C levels in the liver, improving its antioxidant ability. Antistress property of CB was evidenced by swim endurance stress, anorexic stress and despair swim test in rats (Deore and Khadabadi 2009). Dietary inclusion of *Chlorophytum borivilianum* root powder mixed with concentrate at 40 to 80 mg/kg BW/day has been beneficial in enhancing cows' immunity, reducing stress hormones, optimizing plasma metabolites levels and reducing levels of pro-inflammatory cytokines (Devi *et al.* 2021a). Further, it enhanced the production performance of Karan Fries cows in terms of milk yield and quality (Devi *et al.* 2021b).

Conclusion

Heat stress negatively impacts livestock comfort, dry matter intake and subsequently production and reproduction. It is critical to use heat stress ameliorative techniques to prevent production losses from livestock. Reducing the fiber content of the diet will help to reduce metabolic heat production and ultimately improve the heat exchange and reduce body temperature. As feed intake is dramatically reduced under heat stress, a common technique is to enhance the energy density of the diet (reduced fiber, increased concentrates and supplementary fat). The ration should be balanced properly and the concentration of all other nutrients in the diet should be increased. Feed

supplements/additives like, minerals, vitamins and other antioxidants (Melatonin, Astaxanthin, Betaine, and *Chlorophytum borivilianum*) are useful in improving immunity, lowering stress markers and improving productivity. By amelioration of adverse effect of heat stress in dairy animals can improve production performance of animal viz., milk yield and composition, hormonal balance, semen and ovum quality, improved fertility rate and reduced embryonic losses.

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