

ADAPTIVE PHYSIOLOGICAL AND BIOCHEMICAL RESPONSES OF DAIRY ANIMALS TO HEAT STRESS: A REVIEW

MEHUL D. PATEL¹, JIGAR H. PATEL², MAHIPATSIH B. RAJPUT³ & AKSHAY R. BARIYA⁴

^{1,2,3}Lecturer, Kankuba Pashupalan Vidyapith, Institute of Dairy Sciences, Ganpat University, Gujarat, India

⁴M.V.Sc Scholar, Dept. of Livestock Products and Technology, College of Veterinary Science and Animal Husbandry,
Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Gujarat, India

ABSTRACT

Stress is reaction of body to stimuli that disturb homeostasis and adversely affects the animal body system. To cope up such situation animal body shows many neuro-endocrine responses which may alter the biochemical reactions to acclimatize to its surrounding. Heat stress is important phenomenon among dairy farmers due to its deleterious effect on productivity of animal and so economy of farmers. Animal's responses to atmospheric temperature above thermoregulatory critical limits are indicative of animal's adaptation. However, when homeostasis mechanism is not quite enough for thermal adjustment, animal undergoes a critical condition. Better manage-mental practices can assist to minimise heat stress on animal body and to maximize its productivity.

KEYWORDS: Heat Stress, Homeotherms, Panting, Thermoregulation

INTRODUCTION

The physiological responses, biochemical reactions and body temperature are inter-correlated. A rise of temperature accelerates the biochemical reactions, and a lowering of temperature depresses these reactions, likewise appropriate and physiological response is exhibited. Furthermore, it is well known that fluctuations in body temperature are directly correlated with the fluctuation in environmental temperature surrounding the animal as response of animal body to its surrounding temperature whether too cold or too hot. This is physiological response or adjustment to change in external environment has highly effect on productivity of domestic animals. Mammals and birds have developed a thermoregulatory mechanism whereby body temperature is maintained at a relatively constant level regardless of the temperature of the surroundings. Therefore, mammals and birds are classified as homeotherms, or warm-blooded animals. Poikilotherm (cold-blooded) animals have a body temperature that varies with the temperature of the environment.

Homeotherms have optimal temperature zones or thermo-neutral zones for production within which animal's body temperature remains relatively constant [1]. However, long term exposure of animal to the atmospheric temperature below or above the thermo-neutral zone may alter the physiological functions of animal and animal may be called as cold or heat stressed, respectively. Stress is reaction of body to stimuli that disturb homeostasis often with detrimental effects [2]. Environmental heat stress is the most detrimental to dairy animals and results in the hindrance of feed consumption [3], decreased milk production [4], and reproductive performance [5]. Long term exposure to high temperature may results in heat stress causes negative balance between the net amount of energy flowing from the animal to its surrounding environment and the amount of heat energy produced by the animal. Nonetheless high producing animals may relatively more prone to be affected with changes in environmental temperature i.e. high yielding Holstein-Friesian cows in first,

second or third parity at the first stage of lactation are the most susceptible: their heat production is twice that of low yielding or dry cows [6]. Major factors affecting a cow's susceptibility to heat stress are presented in table 1. Besides this, a highly integrated mechanism of behavioural and physiological responses is set in animal which helps to maintain constant internal environment and physiological equilibrium [7, 8].

Table 1: Major Factors Affecting a Cow's Susceptibility to Heat Stress [6]

Breed (imported Holstein-Frisian versus local indigenous breed)	Body condition score (high versus low)
Parity (younger versus older)	Behaviour (active/aggressive versus quiet)
Level of milk production (high versus low)	Housing conditions (well versus poorly ventilated)
Lactation stage (early versus late or dry)	Pasturing conditions (shady versus non-shady)
Level of feed intake (high versus low)	Quality of feedstuffs in the ration
Ration composition (protein rich versus poor)	

PRINCIPLES OF THERMOREGULATION

The basic thermoregulatory approach of a mammal is to maintain a body core temperature higher than ambient temperature to allow heat to flow out from the core via 4 basic routes of heat exchange (conduction, convection, radiation, and evaporation) [9]. Three of these routes (conduction, convection, and radiation) are referred to as sensible routes of heat loss and require a thermal gradient to operate. The fourth (evaporation) works on a vapor/pressure gradient and is defined as insensible heat loss. When ambient temperature conditions approach body temperature, the only viable route of heat loss is evaporation; if ambient conditions exceed body temperature, heat flow will reverse and the animal becomes a heat sink. These all phenomenon directly related to the physiological response of an individual. Therefore, estimating the thermal environment around animals is key to understanding their cooling needs [9].

Thermoregulation is type of negative feedback of homeostasis. Sheep and cattle maintain up their body temperatures within narrow limits over a wide range of atmospheric temperatures by balancing heat loss or gain, and heat production [10]. Metabolic heat and heat from the environment can increase body temperature [11]. Difference in thyroxin activity was observed during hot and cold environmental temperature which is in a straight line with the metabolic heat production [12]. As soon as ambient temperature rises, heat is dissipated primarily by passive mechanisms (sensible heat loss) such as radiation and convection [13]. As ambient temperature approaches skin temperature, the rate of heat dissipation through sensible heat loss decreases. As heat stress progresses, there is recruitment of evaporative processes, primarily sweating and increased respiratory rate [13, 14].

PHYSIOLOGICAL AND BIOCHEMICAL RESPONSES

Different physiological responses involve in thermoregulation. Sweating, high respiration rate, vasodilatation with increased blood flow to skin surface, reduced metabolic rate, decreased dry matter intake and altered water metabolism etc are the physiological responses that have negative impact on the production and reproduction of the cows [15].

Heat is detected by thermo receptors in the skin and buccal membranes, which detect ambient temperature, and also by central receptors in the hypothalamus and spinal cord that can detect changes in the blood temperature [16].

Changes in core body temperature cause the hypothalamus to send nerve impulses to the sweat glands, muscles and blood vessels to raise or lower the temperature. If the core temperature goes up the body loses heat to bring it down again. If the core temperature goes down the body will conserve and even generate heat to bring it up again. Change in rectal temperature has also been considered an indicator of heat storage in animal's body and may be used to assess the adversity of thermal environment, which can affect growth, lactation and reproduction of dairy animals [17, 18].

The initial physiological responses initiated to increase heat loss redirect blood to the periphery, by vasodilatation of skin blood vessels, and vasoconstriction of vessels supplying internal organs. Increased sweating rate then enhances heat loss from the skin. There will be loss of electrolytes in the sweat, with the amount lost differing between species, and perhaps between breeds, with other animal factors such as acclimatisation to heat also influencing the composition. Cattle sweat is hypotonic, containing mostly potassium (K^+), along with sodium (Na^+) and chloride (Cl^-) [15].

If the loss of heat from the skin is not sufficient to maintain stable core temperature, additional heat can be lost from the respiratory membranes as the animal pants. *Panting* is thought to be a more important heat loss mechanism in sheep than cattle [19]. An increased respiration rate is the first reaction when animals were exposed to environmental temperature above the thermo neutral zone [17]. Panting is of particular importance when the humidity increases along with the temperature, as experienced by animals shipped through equatorial regions during the summer. Evaporation of water requires a vapour pressure gradient for loss of heat energy as water evaporates to the surrounding air, but in very humid conditions this gradient is reduced, and therefore evaporative heat loss from the skin is reduced. Respiratory cooling can still occur under these conditions because inspired air is warmed to body temperature and therefore can take on more water vapour, which maintains the gradient. However, when the temperature of inspired air rises to near body temperature, this means of heat loss from panting also becomes limited [20]. Moreover, panting along with sweating generate metabolic heat, imposing an additional heat load [21]. Generally, for animals that both sweat and pant such as cattle, there will be a lag of two to three hours before the respiratory mechanisms for heat loss are initiated. These physiological responses are reviewed in Sparke *et al.* [20]. Evaporation from the skin provides the greater heat loss for cattle, up to about 80% of total evaporative heat loss [22], but panting is an important mechanism for additional heat loss.

Increase in respiratory frequency may be used an index of discomfort in large animals. Al-Haidary [23] also observed significantly higher respiration rate and heart rate in heat stressed sheep than control animal. With increasing environmental temperature, the respiratory rate increases, but the tidal volume decreases. This allows for increased heat loss from the respiratory membranes, but does not increase alveolar ventilation substantially, and therefore does not alter the total gas exchange for the animal. When heat stress becomes more severe, the depth of respiration increases back to near normal tidal volume while the respiratory rate remains elevated above normal (second stage panting). Overall, there is increased airflow over surfaces for evaporative heat loss, but there is also increased alveolar ventilation, up to five times normal in sheep and cattle. The increase in alveolar ventilation leads to excessive expiration of carbon dioxide and respiratory alkalosis [20, 24]. Bhatnagar and Choudhary [25] found that the combination of relative humidity and air temperature caused variation in body temperature and respiration rate of animals, whereas the relative humidity caused variation in pulse rate. McLean [26] found that the significance of increase in respiration rate under heat stress enabled the animal to dissipate the excess body heat by vaporizing more moisture in the expired air, and accounts for about 30 percent of the total heat dissipation. Salem [27] reported an increase in respiration rate if buffaloes and crossbred cattle during summer compared to other seasons. Nonetheless, a higher respiration rate of 71.5/minute during summer compared to

38.8/minute in winter was recorded in lactating cows by Taneja [28].

Sustained panting may be limited by the alkalosis that develops if there is a conflict between the drive to conserve expired carbon dioxide, and the continued elimination of heat through panting. However, buffering mechanisms will operate to maintain blood pH within the normal range [29], and this should allow panting to continue. Mechanisms to restore blood pH involve the immediate release of hydrogen ions (H^+) from intracellular locations, and renal mechanisms that conserve H^+ and excrete bicarbonate ions (HCO_3^-).

Alkalosis does have serious consequences for the well-being of the animal. It is associated with altered neurological function, because a lowered partial pressure of carbon dioxide in the cerebral blood vessels can cause vasoconstriction and reduce perfusion of the brain [30]. Alkalosis can also decrease the availability of minerals such as magnesium and calcium, which can contribute to muscle tremors and tetany.

The change to second stage panting is thought to occur because there is continued heat stimulation of the respiratory centres via the hypothalamus, the carotid bodies, pulmonary receptors or medulla oblongata, along with a reversal of the effect of hypomania on respiration in hyperthermia [24]. The stimulation will be of both rate and depth, but due to the interdependence of the two parameters, increasing the depth of respiration results in a slight decrease in rate.

The impact of these physiological responses on electrolytes will be through the loss of fluid and electrolytes in sweat, and through interactions with the buffering of respiratory alkalosis. Potassium (K^+) and sodium (Na^+) ions are the major cations involved in maintaining acid-base status, and in alkalosis K^+ will exchange with H^+ and enter cells to maintain electro neutrality. Potassium ions will also exchange with H^+ in the renal tubules, and along with sweat losses, this urinary loss can lead to a total body deficit of K. Na^+ is conserved as much as possible in the animal, being the major cations involved in water balance, but when there is low body potassium, there is less aldosterone released, and the main drive for reabsorption of Na^+ from the urine is reduced. This can lead to further Na^+ losses. In situations of depletion of both K^+ and Na^+ , there is less cation available for exchange with H^+ in the urine, and paradoxical aciduria can result, exacerbating the alkalosis.

An important aspect to maintaining normothermia is the reduction in heat production by the animal. This occurs with a decrease in metabolic rate [20]. There will be behavioural responses, which decrease the activity and metabolic rate of the animal. In most animals, hot conditions result in a decreased feed intake, but the mechanism for this is unknown. It could be due to a reduction in the rate of passage of digesta, which increases gut fill for longer and depresses intake. There may also be a direct effect of the increased temperature on the feeding centre of the hypothalamus, resulting in a hormonal response, which could also decrease metabolic rate [31]. Thyroid activity is reduced in situations of heat stress, but the effect of heat on thyroid function takes at least 60 hours to be significant, so this is not an immediate response to acute heat stress, and instead can be involved in the acclimatisation of animals to sustained heat load. A decrease in thyroid hormones will act to decrease the metabolic rate, and reduce the amount of heat produced by the cells. There is some indication of intrinsic species and breed differences in resting metabolic rate that might account for different tolerance to heat.

Other hormonal responses to heat stress include a rise in plasma cortisol with short term exposure to heat, perhaps due to the 'stress' reaction, while in long term heat exposure there is a reduced cortisol turnover rate and a decreased plasma concentration. Growth hormone is also decreased in both short and long term exposure to heat, which will contribute to a lower metabolic rate, but also has implications for growth and production, by animals [31].

There will be behavioural responses to the increased temperature. Animals can change posture, e.g. stand or spread out to increase surface area for heat loss, reduce activity, and seek shade if outside [16].

ADVERSE EFFECTS OF HEAT STRESS

Thermal stress affects almost all systems of body [32]. Many serious effects were also been investigated with heat stress which adversely affect the reproduction of animal and thereby loss of productivity and economic loss. Heat stress reduces libido by reducing level of testosterone, sperm output, decreasing sperm motility and by increasing up proportion of morphologically abnormal spermatozoa in the ejaculate [33]. In female, it lowers fertility, conception rate and embryonic survival in animals [34]. Impairment of follicular and oocyte development as well as reduction in steroid production were also noted [35, 36]. Primary effect of environmental stress in neonates is increased incidence of disease associated with decrease in immunity by reducing immunoglobulin content in plasma. It also reduces fetal growth and alters endocrine status of the dam. Carryover effects of heat stress during late gestation on postpartum lactation and reproduction are also detectable [37].

Various kinds of stress including heat and cold stress leads to the production of reactive oxygen species (ROS) such as superoxide, peroxide, hydroxyl radical, singlet oxygen etc. ROS are also generated during normal body functions [38]. Although, low levels of ROS are essential for many biochemical processes but their accumulation due to over production or a decreased antioxidant defence mechanism causes oxidative stress which leads to damage of bio molecules viz. DNA/RNA, proteins and lipid per oxidation of membranes and disruption of normal cell metabolism [39]. Heat stress may lead to over production of transition metal ions (TMI), which can make electron donations to oxygen forming superoxide or H_2O which is further reduced to an extremely reactive OH radical causing *oxidative stress* [40].

Heat stress also alters the cellular activities. At the molecular and cellular levels, temperature beyond the comfort zone reduces the rates of enzymatic reactions, diffusion, and transport and induces the denaturation and misaggregation of proteins. It also slows down progression through cell cycle, inhibit transcription, translation, disrupt cellular cytoskeleton elements and change membrane permeability [41].

Thermal stress is known to alter the homeostatic mechanism of animals resulting in impaired erythropoietin. High environmental temperature increases oxygen consumption of animals by increasing respiration rate as described above. The higher oxygen intake increases the partial pressure of oxygen in blood, decreases erythropoietin which in turn reduces the number of circulating erythrocytes and thus PCV and Hb values [42-44]. Other explanation of decrease in haemoglobin and PCV levels during thermal stress could be increased attack of free radicals on the erythrocyte membrane, which is rich in lipid content, and ultimate lysis of RBC or inadequate nutrient availability for haemoglobin synthesis as the animal consumes less feed or decreases voluntary intake under heat stress. During summer stress a significant depression in PCV may also be due to haemodilution effect where more water is transported into the circulatory system for evaporative cooling [45].

Metabolic regulators are important in elucidating a picture of modulation in physiological mechanisms during stressed conditions and are best assessed by determining the enzymes governing various metabolic reactions in plasma or serum. Metabolic activities of individual are well reflected by levels of these enzymes in serum. Therefore, during stress condition, the enzymatic activities vary from the animal in comfort zone. Evidence showed that alkaline phosphatase (ALP) and lactic dehydrogenase (LDH) activity was significantly reduced in heat stressed animal [46]. Decrease in these

enzymes during heat stress is due to decrease in thyroid activity during heat stress [46]. Serum level of aspartate transaminase (AST) and alanine transaminase (ALT) is helpful in diagnosis of welfare of animals. Serum ALT value found to be increased during heat stress in goats [47]. No significant changes were observed in AST level in goats during heat stress [47, 48].

Blood glucose and total serum cholesterol levels are physiological adaptation mechanisms that can be affected by high ambient temperatures. Greater variations are observed in levels of blood glucose and total serum cholesterol in hot conditions than in the thermo-neutral zone. Further, some researchers reported that hot climatic conditions are responsible to reduce blood glucose and total serum cholesterol levels [48] and some reports are contrary to these [49]. Bahga *et al.* [50] and Ocak *et al.* [51] reported that blood glucose and total cholesterol level decreases during summer season and increases during winter season in goats. Determination of blood parameters may be important in establishing the effect of heat stress. The well estimated decline in total serum cholesterol levels may have a relation with the increase in total body water or the decrease in acetate concentration which is the primary precursor for the synthesis of cholesterol.

Significant decrease in total protein concentration has been accounted during heat stress [52]. The total plasma protein, albumin, globulin decreased from 6.56, 2.53, 4.04 to 5.88, 2.08 and 3.80 g/dl respectively in balady goats subjected to short term heat stress for two days [46]. This may be due to increase in plasma volume as a result of heat shock which causes results in decreases plasma protein concentration. Prolong exposure of solar radiations was found to elevate the plasma level of total protein, albumin, and globulin. This might be due to vasoconstriction and decreased plasma volume during heat stress [46].

Nevertheless, many researchers stated that heat stress also affects the milk production and its composition which may results from metabolic response of animal to heat stress. Fat and protein percentages were comparatively higher in cows provided cooling than those were not cooled [53]. Similar findings were also reported by El-Khashab [54] in buffaloes. Dupreez [53] recorded that heat stress reduced butter fat by 20–40%, non-fat solids by 10–20% and total milk protein by 10–20%. However, Singh *et al.* [55] did not notice any significant difference in milk composition (fat, SNF and total solids) between heats stressed and protected Nili-Ravi buffaloes. Moreover, the negative effect of heat stress on milk production is due to the decreased nutrient intake and decreased nutrient uptake by the portal drained viscera of the buffalo [56].

Heat stress adversely affect productivity and reproductively of the animals and thus economy of farmers. Better management reduces heat stress includes nutrient supplementation, microclimate modifications like improving shade, air movement, cooling apparatus, water intake, night grazing, feeding high energy diet as well as proper supervision help reduce heat stress. It has beneficial effect on physiological responses, total milk production and compositions in lactating buffaloes during hot dry and hot humid summer months.

CONCLUSIONS

Thermal stress is a cause of great concern among livestock owners in tropical countries. In hot-humid climates, although animal attempts to acclimatize through physiological changes including cutting down on feed intake and heat production, but this does not come without sacrificing part of its productivity. Heat stress causes change in the antioxidant level and various hormones and electrolyte concentrations. Animal illustrates various responses to thermal stress at physiological, cellular, biochemical, molecular and systemic level that collectively affect the animal productivity and

economy of farmers. In order to prevent this economic loss to the farmer, there is need to understand and effectively combat heat stress by minimizing its impact on animal body and its productivity.

REFERENCES

1. Farooq, U., Samad, H., Shehzad, F. and Qayyam, A. (2010). Physiological responses of cattle to heat stress. *World Applied Sciences Journal*, **8**:38-43.
2. David, K., Murgo, A.J. and Faith, R.E. (1990). Effects of stress on the immune system. *Immunology Today*, **11**:170-179.
3. Bernabucci, U., Bani, P., Lacetera, R.N. and Nardone, A. (1999). Influence of short and long term exposure to a hot environment on rumen passage rate and diet digestibility by Friestian heifers. *Journal of Dairy Sciences*, **82**:967-973.
4. Sharma, A.K., Rodriguez, L.A., Wilcox, C.J., Collier, R.J., Bachman, K.C. and Martin, F.G. (1988). Interactions of climatic factors affecting milk yield and composition. *Journal of Dairy Sciences*, **71**:819-825.
5. Cavestany, D., El-Whisky, A.B. and Foot, R.H. (1985). Effect of season and high environmental temperature on fertility of Holstein cattle. *Journal of Dairy Science*, **68**:1471-1478.
6. Noordhuizen, J. and Bonnefoy, J. (2015). Heat stress in dairy cattle: Major effects and practical management measures for prevention and control. *SOJ Veterinary Sciences*, **1**(1):1-7.
7. Johnson, H.D. (1985). Physiological responses and productivity of cattle. Chapter 1 in "Stress Physiology in Livestock Volume II Ungulates". Ed M.K. Yousef. CRC Press USA. Pp 3-24.
8. Kamal, T.H., (1965). Physiological reactions of cows to hot environmental conditions. Proc. Symp. On Use of Radioisotopes in Animal Nutrition and Physiology (Prague 23-27 Nov. 1964). Proc. Series IAEA. Vienna. 767
9. Collier, R.J., Dahl, G.E. and VanBaale, M.J. (2006). Major advances associated with environmental effects on dairy cattle. *Journal of Dairy Science*, **89**(4):1244-1253.
10. Crawshaw, L.J. (1980). Temperature regulation in vertebrates. *Annual Review of Physiology*, **42**:473-491.
11. Yousef, M.K. and Johnson, H.D. (1985) Body fluids and thermal environment. *Stress Physiology in Livestock*, vol. I. Basic Principles, CRC Press, Boca Raton, FL, USA, pp. 133-141.
12. Das, K.S., Singh, K., Singh, G., Upadhyay, R.C., Malik, R. and Oberoi, P.S. (2014). Heat stress alleviation in lactating buffaloes: Effect on physiological response, metabolic hormone, milk production and composition. *Indian Journal of animal Sciences*, **84**(3):275-280.
13. Caufield, M., Cambridge, H., Foster, S.F. and McGreevy, P.D. (2014). Heat stress: a major contributor to poor animal welfare associated with long-haul live export voyages, **192**(2):223-228.
14. Mortola, J.P. and Frappell, P.B. (2000). Ventilatory responses to changes in temperature in mammals and other vertebrates. *Annual Review of Physiology*, **62**:847-874.
15. West, J.W. (1999). Nutritional strategies for managing the heat stresses dairy cow. *Journal of Animal Sciences*, **77**:21-35.

16. Bligh, J.A. (1985). Temperature regulation. Chapter 8 in “Stress Physiology in Livestock Volume I Basic Principles”. Ed M.K. Yousef. CRC Press USA. Pp75-96.
17. Ganaie, A.H., Shanker, G., Bumla, N.A., Ghasura, R.S., Mir, N.A., Wani, S.A. and Dudhatra, G.B. (2013). Biochemical and physiological changes during thermal stress in bovines. *Journal of Veterinary Science and Technology*, **4**:126.
18. West, J.W., Hill, G.M., Fernandez, J.M., Mandebvu, P., Mullinix, B.G. (1999). Effects of dietary fiber on intake, milk yield, and digestion by lactating dairy cows during cool or hot, humid weather. *Journal of Dairy Science*, **82**:2455-2465.
19. Thwaites, C.J. (1985). Physiological response and productivity in sheep. Stress Physiology in Livestock, vol. II. Ungulates, CRC Press, Boca Raton, FL, USA, pp. 25–38.
20. Sparke, E.J., Young, B.A., Gaughan, J.B., Holt, S.M., and Goodwin, P.J. (2001). Heat load in feedlot cattle. Project Number FLOT.307, 308, 309. Meat and Livestock Australia Ltd.
21. Bianca, W. (1965). Reviews of the progress of dairy science. Section A. Physiology. Cattle in a hot environment. *Journal of Dairy Research*, **32**:291
22. Robertshaw, D. (1985). Heat loss of cattle. Chapter 6 in “Stress Physiology in Livestock Volume I Basic Principles”. Ed M.K. Yousef. CRC Press USA. Pp55-66.
23. Al-Haidary, A.A. (2004). Physiological response of Naimey sheep to heat change challenge under semi-arid environments. *International Journal of Agriculture and Biology*. **6**(2):307-309.
24. Hales, J.R.S. (1976). Interactions between respiratory and thermoregulatory systems of domestic animals in hot environments. Progress in Biometeorology. Ser. B. *Animal Biometeorology*. **1**(1): 123-131.
25. Bhatnagar, D.s. and Choudhary, N.C. (1960). Influence of climate on the physiological reactions in Murrah buffalo calves. *Indian Veterinary Journal*, **37**:404-409.
26. McLean, J.A. (1963). The regional distribution of cutaneous moisture vaporization in the Ayrshire calf. *Journal of Agricultural Sciences*, **61**:275-280.
27. Salem, I.A. (1980). Seasonal variations in some body reactions and blood constituents in lactating buffaloes and friestian cows with reference to acclimatization. *Journal of the Egyptian Veterinary Medical Association*. **40**:63-72.
28. Taneja, G.C. (1960). Effect of hot environment on rectal temperature, respiratory frequency and respiratory volume of calves. *Indian Journal of Veterinary Sciences and A.H.* **30**:107-113.
29. Whitehair, K.J., Haskins, S.C., Whitehair, J.G. and Pascoe, P.J. (1995). Clinical applications of quantitative acid-base chemistry. *Journal of Veterinary Internal Medicine*, **9**:1-11.
30. Adroque, H.J. and Madias, N.E. (1998). Management of life-threatening acid-base disorders. Second of two parts. *New England Journal of Medicine*, **338**: 107-111.
31. Johnson, H.D. (1965). Response of animals to heat. Meteor. Monogr, **6**:109

32. Gupta, M., Kumar, S., Dangi, S.S. and Jangir, B. (2013). Physiological, Biochemical and Molecular Responses to Thermal Stress in Goats. *International Journal of Livestock Research*, **3**(2): 27-38.
33. Perez-Crespo, M., Pintado, B. and Gutierrez-Adan, A. (2008). Scrotal heat stress effects on sperm viability, sperm DNA integrity, and the offspring sex ratio in mice. *Molecular Reproduction and Development*. **75**:40-47.
34. Sakatani, M, Kobayashi, S. and Takahashi, M. (2004). Effects of heat shock on in vitro development and intracellular oxidative state of bovine preimplantation embryos. *Molecular Reproduction and Development*, **67**:77-82.
35. Zeron, Y, Ocheretny, A., Kedar, O., Borochoy, A., Sklan, D. and Arav A. (2001). Seasonal changes in bovine fertility: relation to developmental competence of oocytes, membrane properties and fatty acid composition of follicles. *Reproduction*. **121**:447-454.
36. Ozawa, M., Tabayashi, D., Latief, T.A., Shimizu, T., Oshima, I. and Kanai, Y. (2005). Alterations in follicular dynamics and steroidogenic abilities induced by heat stress during follicular recruitment in goats. *Reproduction*. **129**(5): 621-630.
37. Collier, R.J., Beede, D.K., Thatcher, W.W., Israel, L.A. and Wilcox, L.S. (1982). Influences of environment and its modification on dairy animal health and production. *Journal of Dairy Science*, **65**:2213-2227.
38. Boveris, A. and Chance, B. (1973). The mitochondrial generation of hydrogen peroxide. General properties and effect of hyperbaric oxygen. *Biochemical Journal*. **134**(3):707-716.
39. Spurlock, M.E. and Savage, J.E. (1993). Effects of dietary protein and selected antioxidants on fatty haemorrhagic syndrome induced in Japanese quails. *Poultry Science*. **72**:2095-2105.
40. Agarwal, A. and Prabhakaran, S.A. (2005). Mechanism, measurement and prevention of oxidative stress in male reproductive physiology. *Indian Journal of Experimental Biology*. **43**:963-974.
41. Sonna, L.A., Fujita, J., Gaffin, S.L. and Lilly, C.M. (2002). Invited review: Effects of heat and cold stress on mammalian gene expression. *Journal of Applied Physiology*. **92**:1725- 1742.
42. Sivakumar, A.V.N., Singh, G. and Varshney, V.P. (2010). Antioxidants supplementation on acid base balance during heat stress in goats. *Asian-Australian Journal of Animal Science*, **23**(11):1462-1468.
43. Maurya, V.P., Naqvi, S.M.K., Joshi, A. and Mittal, J.P. (2007). Effect of high temperature stress on physiological responses of Malpura Sheep. *Indian Journal of Animal Science*. **77**:1244-1247.
44. Kumar, M., Jindal, R. and Nayyar, S. (2011). Influence of heat stress on antioxidant status in beetal goats. *Indian Journal of Small Ruminant*. **17**(2):178-181.
45. EL-Nouty, F.D., Al-Haidary, A.A. and Salah, M.S. (1990). Seasonal variations in hematological values of high and average-yielding Holstein cattle in semi-arid environment. K. S. U. *Agricultural Science*. **2**:172-173.
46. Helal, A, Hashem, A.L.S., Abdel-Fattah, M.S. and El-Shaer. (2010). Effects of heat stress on coat characteristics and physiological responses of balady and Damascus goat in Sinai Egypt. *American Euresian Journal of Agriculture & Environmental Science*. **7**(1):60-69.

47. Sharma, A.K. and Kataria, N. (2011). Effects of extreme hot climate on liver and serum enzymes in marwari goat. *Indian Journal of Animal Science*. **81**(3):293-295.
48. Ocak, S., Darcan, N., Cankaya, S. and Cinal, T. (2009). Physiological and biochemical responses in German Fawn kids subjected to cooling treatments under mediterranean climate conditions. *Turkish Journal of Veterinary and Animal Science*. **33**(6):455-461.
49. Webster, A.J.F. (1976). The influence of the climatic environment on metabolism on cattle. In: Swan, H. and Broster, W.H.(eds), *Principles of Cattle Production*, Butterworths, London.
50. Bahga, C.S., Sikka, S.S. and Saijpal, S. (2009). Effect of seasonal stress on growth rate and serum enzyme levels in young crossbred calves. *Indian Journal of Animal Research*. **43**(4):288-290.
51. Ocak, S. and Guey, O. (2010). Physiological responses and some blood parameters of bucks under Mediterranean climate conditions. *Anadolu Journal of Agricultural Science*. **25**(2):113-119.
52. Dangi, S.S., Gupta, M., Maurya, D., Yadav, V.P., Panda, R.P., Singh, G., Mohan, N.H., Bhure, S.K., Das, B.C., Bag, S., Mahapatra, R.K. and Sarkar, M. (2012). Expression Profile of HSP genes during different seasons in goats (*Capra hircus*). *Tropical Animal Health and Production*. **44**:1905–1912.
53. Dupreez, J.H. (1994). 'The role of heat stress in the performance of dairy cattle in southern Africa.' Ph.D. Thesis, University of Pretoria.
54. El-Khashab, M.A. (2010). Physiological and productive responses to amelioration of heat stress in lactating buffaloes under hot summer conditions in Egypt. Available at: <http://www.spsa-egy.org/?p=835>
55. Singh G, Kamboj M L and Patil N V. (2005). Effect of thermal protective measures during hot humid season on productive and reproductive performance of Nili-Ravi buffaloes. *Indian Buffalo Journal*, **3**:101–04
56. Balhara, A.K., Jerome, A. and Singh, I. (2015). Buffalo under Heat Stress. Available at: http://www.buffalopedia.cirb.res.in/index.php?option=com_content&view=article&id=327&Itemid=255&lang=en