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Influence of ambient temperatures on metabolic responses of Murrah buffaloes of varying physiological states from arid tracts in India

Ashish Joshi[1], Nalini Kataria[1], Anil Kumar Kataria[1], Nidhi Pandey[1], Laxmi Narayan Sankhala[1], Shesh Asopa[2], Ruchi Pachaury[3], Shokat Khan[3]

[1] Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan, India;
[2] Apollo College of Veterinary Medicine, Jaipur, Rajasthan, India;
[3] Government of Rajasthan, India;
Corresponding author: N. Kataria, nalinikataria@rediffmail.com

Abstract. Ambient temperature associated variations in metabolic responses of Murrah breed of buffaloes of varying physiological states belonging to arid tracts were investigated. Healthy adult female Murrah buffaloes were grouped according to physiological states into group A (non-pregnant milch, pregnant milch and pregnant dry) and group B (primipara and multipara) and blood samples were collected during moderate, hot and cold environmental temperature periods to obtain sera. Metabolic responses were assessed by analyzing serum urea, creatinine, total proteins, cholesterol, triglycerides and glucose and the moderate mean values were 5.18±0.04 mmol L^{-1}, 118.00±1.00 µmol L-1, 71.00±0.12 gL^{-1}, 3.30±0.02 mmol L^{-1}, 1.28±0.01 mmol L^{-1} and 3.7±0.02 mmol L^{-1}, respectively. The mean values of serum urea and creatinine were significantly ($p \leq 0.05$) higher during hot and cold ambiences, the mean value of total serum proteins and triglycerides were significantly ($p \leq 0.05$) lower during hot and cold ambiences and the mean values of cholesterol and glucose were significantly ($p \leq 0.05$) lower during hot ambience and significantly ($p \leq 0.05$) higher during cold ambience as compared to respective moderate mean value. The mean values of non pregnant milch, pregnant milch and pregnant dry animals differed significantly ($p \leq 0.05$) from each other in all the environmental periods. It was concluded that extreme hot and cold ambient temperatures affected the buffaloes of all physiological states which was evident in the form of variations in the metabolic responses to combat the environmental challenges.
Key words: Ambient temperature, cold, hot, Murrah buffalo, metabolic responses.

Introduction. Maintenance of anequilibrium between the heat production and heat loss is a prerequisite for thermoregulation in mammals. When thermoneutral environmental conditions are changed to adverse conditions, large domestic animals become stressed which can be observed in the form of modulations in the physiological mechanisms. Since the heat tolerance capacity of buffalo is poor, exposure to heat stress may bring considerable changes in physiological mechanism (Chaiyabutr et al 1987). During drastic change in ambient temperatures, maintenance of health of an animal depends upon the synergistic approach of various body systems. Environmental stress is considered as a big financial burden to animal owners. To cope up with changing environmental temperatures, metabolic adjustments are required.

Diversion of metabolic responses in thermoregulation of an animal may impose great risk to productivity because latter needs nutrients at a large scale. Such situations may produce a threat to health also. An understanding of the extent of modulations in metabolic responses is essential to formulate the ration and to maintain animals in good health. Much stress has been given to heat stress related variations in physiological reactions of the animals. Now the scientific community has shown attention about the thermal comfort of milch animals belonging not only to tropical regions but also of temperate areas (Nardone et al 2010). Importance of Murrah breed of buffalo in milk production is well known. Owing to paucity of literature on metabolic responses of Murrah buffaloes with changing ambient temperatures, the present investigation was launched.

The data generated regarding various physiological states will help in healthy management of these animals and will contribute to future research.

Materials and Methods

Animals and sampling. To find out variations in metabolic responses associated with environmental temperatures, four hundred and fifty apparently healthy adult female Murrah buffaloes between 4 and 12 years of age were screened. Blood samples were collected from jugular vein to harvest the serum in clean and dry test tubes during moderate (mean maximum ambient temperature 30.33±0.20), hot (mean maximum ambient temperature 45.5±0.08) and cold (mean minimum ambient temperature 4.88± 0.20) ambiences. All the animals belonged to private dairy farms in Rajasthan state, India and were managed in similar conditions of feeding and watering. In each ambience 150 blood samples were collected during morning hours. On the basis of physiological states, animals were broadly divided into group A (non-pregnant milch, pregnant milch and pregnant dry) and group B (primipara and multipara). Each category consisted of 30 animals in each ambience.

Analysis. Metabolic responses in the serum included urea, creatinine, total proteins, cholesterol, triglycerides and glucose which were determined by the techniques of Natelson (Varley 1988), Bonsnes and Taussky (Varley 1988), Lowry et al (1951), Sackett (Varley 1988), GPO-PAP method of kit (Wipro) and Folin-Wu (Oser 1976), respectively. The changes in the means were measured by using multiple mean comparison procedures (Duncan 1955 and Steel & Torrie 1980).

Results and Discussion. Mean±SEM values of serum urea, creatinine, proteins (Table 1), cholesterol, triglycerides and glucose (Table 2) have been presented according to ambient temperatures with further categorization as per physiological states.

The mean values of serum urea and creatinine were significantly ($p \leq 0.05$) higher during hot and cold ambiences, the mean value of total serum proteins and triglycerides were significantly ($p \leq 0.05$) lower during hot and cold ambiences and the mean values of cholesterol and glucose were significantly ($p \leq 0.05$) lower during hot ambience and significantly ($p \leq 0.05$) higher during cold ambience as compared to respective moderate mean value. In group A, mean values of all serum metabolites of non pregnant milch, pregnant milch and pregnant dry animals differed significantly ($p \leq 0.05$) from each other in all the ambiences. In each ambience the mean value of serum urea, creatinine, total proteins, cholesterol and glucose were highest in non pregnantmilch animals and were lowest in pregnant dry animals. The mean value of serum triglyceride showed the reverse trend. In group B, the mean values of serum urea, creatinine and triglycerides were significantly ($p \leq 0.05$) higher in multipara animals than primipara in each ambience whereas of cholesterol and glucose were significantly ($p \leq 0.05$) higher in primipara. Though effect of physiological states on serum metabolite concentration in buffaloes is documented (Lapitan et al 2008; AbdEllah et al 2010; Serdaru et al 2011), a comprehensive study to discuss them according to varying ambient temperatures is lacking. It has been seen that metabolite concentrations showed interactions among various responses.

Urea. Higher serum urea showed the increased activity of urea cycle thereby causing profound effect during extreme hot and cold conditions (Nazifi et al 2003). Earlier research has thrown some light over the role of urea and possible development of oxidative stress due to extreme ambiences by correlating the urea with the parameters of oxidative stress (Chena et al 2008). This is probably based on the fact that as a part of oxidative stress mediated solute-signaling pathway in tissues, urea increases expression of the oxidative stress-responsive transcription factor and therefore is associated with oxidative stress. Further, antioxidant treatment partially inhibit the ability of urea to activate transcription of reporter gene (Zhang et al 1999). These findings indicates toward the possible role of urea in oxidative stress mechanisms. The status of protein metabolism in the body influences serum urea level. Low levels of serum urea in primipara could be due to comparatively lower protein catabolism in comparison to multipara (Coles 1986).

Table 1

Mean ± SEM values of serum urea, creatinine and total proteins in Murrah buffalo

Effects	Serum metabolic responses		
	Urea mmol L^{-1}	Creatinine umol L^{-1}	Total proteins g L^{-1}
Ambient temperatures			
Moderate (150)	5.18±0.04 [b]	118.00±1.00 [b]	71.00±0.12 [b]
Group A			
Non-pregnant milch (30)	6.62±0.04 [d]	126.00±1.04 [d]	77.00±0.10 [d]
Pregnant milch (30)	4.72±0.03[d]	119.00±1.10[d]	72.00±0.11 [d]
Pregnant dry (30)	4.20±0.04[d]	109.00±1.00[d]	64.00±0.10[d]
Group B			
Primipara (30)	4.07±0.03[f]	107.00±1.20[f]	66.0±0.22 [f]
Multipara (30)	6.29±0.03 [f]	129.00±0.10[f]	76.0±0.18 [f]
Hot (150)	7.17±0.03[b]	215.00±1.10 [b]	61.50±0.20 [b]
Group A			
Non-pregnant milch (40)	8.51±0.04 [d]	221.00±1.10 [d]	66.30±0.20 [d]
Pregnant milch (40)	7.00±0.03[d]	214.00±1.40 [d]	61.80±0.20 [d]
Pregnant dry(40)	6.02±0.03 [d]	200.00±1.80 [d]	56.40±0.22 [d]
Group B			
Primipara (30)	6.19±0.03 [f]	205.00±1.10[f]	55.50±0.20 [f]
Multipara (30)	8.15±0.04 [f]	225.00±1.30 [f]	67.50±0.22 [f]
Cold (150)	5.78±0.03[b]	210.00±1.00 [b]	67.00±0.06 [b]
Group A			
Non-pregnant milch (30)	7.08±0.04 [d]	230.00±1.22 [d]	71.00±0.10 [d]
Pregnant milch (30)	6.18±0.03 [d]	205.00±1.00[d]	68.00±0.15[d]
Pregnant dry(30)	4.08±0.02[d]	195.00 ±1.30[d]	62.00±0.10 [d]
Group B			
Primipara (30)	4.99±0.02 [f]	201.00±1.00 [f]	64.00±0.20 [f]
Multipara (30)	6.57±0.03 [f]	219.00±1.00 [f]	70.00±0.20 [f]

Figures in the parenthesis indicate number of animals. Means superscribed by same superscript within a column differ significantly ($p \leq 0.05$)

Creatinine. Environmental stress can result into higher serum creatinine concentration due to higher metabolic activity in liver and muscle mediated by cortisol (Kataria et al 2000a). In human patients higher serum creatinine levels have been correlated with oxidative stress (Kolagal et al 2009). Due to paucity of such studies in animals, it is very important to link creatinine as one of the important diagnostic parameters of environment related oxidative stress. Heat stress mobilises proteins and produces variations in value (Nath 2006). Metabolic status of the animals is also reflected by the levels of creatinine. Most of the creatinine excreted originates from endogenous creatine. The amino acids arginine and glycine combine to form guanidinoacetate in the pancreas, kidney and small intestine. In the liver, methionine provides a methyl group for conversion of guanidinoacetate to creatine. Creatine circulates in plasma and is taken up by muscle, where it stores energy in the form of phosphocreatine, which undergoes spontaneous cyclisation with loss of inorganic phosphate to form creatinine. Cretaine undergoes no catabolic reaction other than decomposition to creatinine (Finco 1999). The quantity of creatinine formed each day depends also upon the rate of synthesis of creatine by the liver (Kaneko et al 1999) and stress related changes may occur in the rate of formation.

Table 2

Mean ± SEM values of serum cholesterol, triglycerides and glucose in Murrah buffalo

Effects	Serum metabolic responses (mmol L^{-1})		
	Cholesterol	Triglycerides	Glucose
Ambient temperatures			
Moderate (150)	3.30±0.02 [b]	1.28±0.01 [b]	3.70±0.02 [b]
Group A			
Non-pregnant milch (30)	3.90±0.01 [d]	1.15±0.01 [d]	3.81±0.02 [d]
Pregnant milch (30)	3.40±0.01 [d]	1.29±0.01 [d]	3.71±0.01 [d]
Pregnant dry(30)	2.60±0.02 [d]	1.40±0.01 [d]	3.58±0.01 [d]
Group B			
Primipara (30)	3.7±0.02[f]	1.16±0.02 [f]	3.79±0.02[f]
Multipara (30)	2.9±0.02[f]	1.40±0.01 [f]	3.61±0.02[f]
Hot (150)	2.70±0.02[b]	0.40±0.01[b]	3.10±0.02[b]
Group A			
Non-pregnant milch (30)	2.83±0.03 [d]	0.29±0.02 [d]	3.30±0.02[d]
Pregnant milch (30)	2.69±0.02[d]	0.39±0.01 [d]	3.10±0.01[d]
Pregnant dry(30)	2.58±0.03 [d]	0.52±0.01 [d]	2.90±0.01 [d]
Group B			
Primipara (30)	2.81±0.02[f]	0.27±0.01 [f]	2.80±0.02[f]
Multipara (30)	2.59±0.02[f]	0.53±0.02 [f]	3.40±0.01[f]
Cold (150)	4. 7±0.03[b]	0.93±0.01[b]	3.80±0.01[b]
Group A			
Non-pregnant milch (30)	5.8±0.03 [d]	0.83±0.01 [d]	4.10±0.02 [d]
Pregnant milch (30)	4.6±0.02[d]	0.90±0.01[d]	3.79±0.01[d]
Pregnant dry(30)	3.7±0.03 [d]	1.06±0.01 [d]	3.51±0.01[d]
Group B			
Primipara (30)	5.5±0.02[f]	0.76±0.01[f]	4.00±0.02[f]
Multipara (30)	3.9±0.03[f]	1.10±0.02[f]	3.60±0.03[f]

Figures in the parenthesis indicate number of animals. Means superscribed by same superscript within a column differ significantly ($p \leq 0.05$)

Proteins. Stress conditions may produce oxidative changes in proteins leading to high turnover in blood (Goswami et al 2003). It can be hypothesised that total serum proteins can also serve as marker of environment related stress. Reactive oxygen species formed due to extreme variations in the environmental temperatures can modify concentrations of fuel molecules producing oxidative stress as superoxide radicals and hydroxyl radicals have a great impact on the normal function of biomolecules (Nazifi et al 2009). The higher value of total serum proteins in comparatively older age group may be suggestive of increased physiological activity of hepatocytes.

Cholesterol. Higher serum cholesterol levels could be attributed to higher thyroid activity in cold ambience as increase in BMR is required with the need for extra production of heat to maintain body temperature (Kataria et al 2000b). During heat stress liable changes in the phytochemistry of the normal grazing flora could add to the factors accounting for the lowering of the plasma lipids concentration (Al-Qarawi 1999). Recent advances in the field of clinical medicine links the cholesterol with oxidative stress (Pappolla et al 2002). Low cholesterol levels can be related to higher metabolic needs (Saeed et al 2004). Growth and different age groups can affect the metabolite status as per the need of the body (Kataria et al 2000b). Higher level of cholesterol could be due to oestrogen effect which promotes cholesterol synthesis (Singh et al 1994).

Triglyceride. Changes in triglyceride level indicated the metabolic status of the animals during extreme ambiences (Karapehlivan et al 2007). Oxidative stress was correlated with the triglyceride levels by various workers in human subjects (Katsuki et al 2004). The higher values of serum triglycerides could be due to higher metabolic responses to meet the energy requirements and can be related to metabolic stress response (Mazuri et al 2009).

Glucose. Hot and cold conditions function as stressors to the animals whereby increasing glucocorticoid secretion, which induces gluconeogenesis and inhibits peripheral utilisation of glucose, and increases or maintains the blood glucose in stressed animals (Weber et al

1965). During heat stress feed consumption decreases, which comparatively lowers the blood glucose (Kataria et al 2002). The lower blood glucose during hot ambience might also be due to variation in thyroid hormones (Nath 2006). The association between glucose and reactive oxygen species has been discussed widely (Russella et al 2002). Emerging data from human and animal studies suggest that glucose-derived oxidative stress may play a central role, linking together many of the other physio-pathogenetic mechanisms (Greene et al 1999).

Conclusion. It was concluded that variations in the ambient temperature uring extreme hot and cold ambiences affected the buffaloes of all the physiological states. Probably modulations in the metabolic responses reflected towards metabolic adaptation to combat environmental challenges. Present investigationhas attempted to relate the modulations in metabolic responses according to variations in the ambient temperatures in Murrah breed of buffaloes at a large scale. The data generated in the present investigation could help in the future research in the field of metabolic stress responses.

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Authors:
Ashish Joshi, Department of Veterinary Physiology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India.
Nalini Kataria, Department of Veterinary Physiology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India, Phone , 0091-151-2546399; e-mail: nalinikataria@rediffmail.com
Anil Kumar Kataria, Apex Centre for Animal Disease Investigation, Monitoring and Surveillance, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India.
Nidhi Pandey, Department of Veterinary Physiology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India.
Laxmi Narayan Sankhala, Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India
Shesh Asopa, Department of Veterinary Pathology, Apollo College of Veterinary Medicine, Jaipur, Rajasthan, India.
Ruchi Pachaury, Department of Animal Husbandry, Government of Rajasthan, India.
Shokat Khan, Department of Animal Husbandry, Government of Rajasthan, India.

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