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ORIGINAL ARTICLE

Heat stress induced changes in metabolic regulators of donkeys from arid tracts in India

Kataria N.[1*], Kataria A.K.[2]

[1] *Department of Veterinary physiology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334 001, Rajasthan, India*
[2] *Apex Centre for Animal Disease Investigation, Monitoring and Surveillance, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner – 334001, Rajasthan, India*

Phone 0091 – 151- 2546399

[*]*E-mail:**nalinikataria@rediffmail.com***

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To find out heat stress induced changes in metabolic regulators of donkeys from arid tracts in India, blood samples were collected to harvest the serum during moderate and extreme hot ambiences. The metabolic enzymes determined were sorbitol dehydrogenase, malate dehydrogenase, glucose-6-phosphate dehydrogenase, glutamate dehydrogenase, ornithine carbamoyl transferase, gamma-glutamayl transferase, 5'nucleotidase, glucose-6-phosphatase, arginase, and aldolase. The mean values of all the serum enzymes increased significantly ($p \leq 0.05$) during hot ambience as compared to respective values during moderate ambience. It was concluded that increased activity of all the enzymes in the serum was due to modulation of metabolic reactions to combat the effect of hot ambience on the animals. Activation of gluconeogenesis along with hexose monophosphate shunt and urea cycle probably helped the animals to combat the heat stress.

Key words: Donkeys; heat stress; gluconeogenesis; hexose monophosphate shunt; metabolic enzymes

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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 blood or serum. The level of these enzymes reflects the metabolic activities and commonly used by the researchers to find out the metabolic responses. Arid tracts experience the problem of extreme variations in ambient temperatures which makes it essential to determine the adaptability level of the animals living in these areas to increase their sustainability by taking proper and timely measures (Kataria and Kataria, 2006).

Although donkeys are important animals of arid areas but scientific approach in their management is generally ignored in the name of they being hardy animals. However, it is important to protect them from harsh climatic changes. Extreme hot ambience causes significant stress with the disturbance in the physiological functioning (Kataria and Kataria, 2010). The profile of enzymes governing metabolic

reactions in the serum can help in substantiating the clinical examination providing an excellent basis for correctly diagnosing the stress condition. It is important to note that in addition to the pathological conditions, various kinds of stressors are able to make considerable changes in the physiological reactions related with metabolism. This could either be due to increased production or by increased leakage through the cell membranes. Hence, it becomes important for every laboratory to establish reference values of the parameters of native animals. Monitoring of the biochemical mechanisms during ambient stresses can help in the health management of the animals.

The diagnosis of disease is mainly dependent upon deviations from the normal range of physiological values. However, the absence of established reference values may lead to misinterpretation of diagnosis and treatment of different disease conditions. To determine the diagnostic value of the blood serum level of the enzyme, it is necessary to carry out investigations in stressed animals when affected with various diseases and then compare them with the normal range of values given for those animals, preferably of the same breed, and in similar environmental conditions. Looking towards the paucity of literature on the ambience affected changes in metabolic regulators of donkeys from arid tract, the present investigation was planned to determine variations in serum enzymes governing key metabolic reactions in healthy donkeys and to provide normal reference values for future use in disease diagnosis.

MATERIALS AND METHODS

To find out the changes in metabolic regulators due to heat stress, enzymes of metabolic reactions in serum were determined in donkeys found in the arid tracts of Rajasthan state, India. The animals were maintained in the similar management and feeding conditions. Blood samples were collected during moderate ambience (maximum temperature of 28^0C- 29^0C) and hot ambience (maximum temperature of 44^0C- 46.5^0C) from 30 same adult male animals which were free from endo-parasites as assessed by routine faecal examination.

The enzymes of metabolic reactions in serum were sorbitol dehydrogenase (SDH), malate dehydrogenase (MDH), glucose-6-phosphate dehydrogenase (G6PDH), glutamate dehydrogenase (GDH), ornithine carbamoyl transferase (OCT), gamma-glutamayl transferase (γGT), 5'nucleotidase (5'NT), glucose-6-phosphatase (G-6-Pase), arginase (ARG) and aldolase (ALD). Serum enzyme activities were measured according to the specific reaction of each enzyme by using basic standard techniques as described by King (1965) for SDH, MDH, G6PDH, GDH and G-6- Pase; by Brown and Grisolia (1959) for OCT; by Wolf and William (1973) for γGT; by Varley (1988) for 5'NT; by Manning and Grisolia (1957) for ARG and by Sibley and Lehninger (1949) for ALD. All results of enzyme activities were expressed as per SI units in Units/litre written as UL^{-1}. Statistical significance for individual parameter between healthy and heat stressed group was analysed as per Snedecor and Cochran (1967).

RESULTS AND DISCUSSION

The mean values of enzymes of metabolic reactions in serum are presented in table 1. In heat stressed group the mean values of all the enzymes increased significantly ($p \leq 0.05$) as compared to respective moderate mean value.

Table 1. Mean ± SEM values of serum enzymes in donkeys of arid tract

Serum enzymes, UL^{-1}	Moderate ambience (30)	Hot ambience (30)
Sorbitol dehydrogenase	11.66±0.11	33.42±2.20 [b]
Malate dehydrogenase	40.22±1.11	80.13±1.40 [b]
Glucose- 6- phosphate dehydrogenase	3.98±0.01	9.12±0.09 [b]
Glutamate dehydrogenase	24.33±0.89	48.21±3.03 [b]
Ornithine carbamoyl transferase	12.00±1.00	39.46±2.44 [b]
Gamma glutamyl transferase	25.15±1.13	56.15±3.03 [b]
5' nucleotidase	16.44±1.11	48.15±3.03 [b]
Glucose- 6- phosphatase	2.99±0.09	8.17±3.16 [b]
Arginase	10.51±0.07	30.43±2.11 [b]
Aldolase	10.22±0.09	30.14±2.10 [b]

Figures in the parenthesis indicate number of animals.
Superscript 'b' indicates a significant difference ($p \leq 0.05$) from respective moderate mean value.

The increased activity of sorbitol dehydrogenase during hot ambience probably helped in meeting the demand for glucose (Wolf and Williams,1973) as it is an enzyme of carbohydrate metabolism converting sorbitol into fructose and with aldose reductase, it provides a way for the body to produce fructose from glucose without using ATP. The higher activity also suggested stress mediated leakage (Alemu *et al.,* 1977). Malate dehydrogenase is an enzyme in the citric acid cycle that catalyzes the conversion of malate into oxaloacetate and *vice versa*. It is also involved in gluconeogenesis. Higher activity of MDH in hot ambience indicated the strategies of the animal to modulate the metabolic pathways for energy generation (Kataria *et al*., 2011). Glucose-6-phosphate dehydrogenase is an enzyme of pentose phosphate pathway, a metabolic pathway which supplies reducing energy to the cell. Increased activity of G6PDH during extreme hot ambience showed the liver stimulation. This enzyme is highly active in liver than other tissues (Hanson and Ballard, 1967). Higher activity of this enzyme reflected that PPP was operative actively in heat stressed animals.

Higher activity of glutamate dehydrogenase suggested increased fuelling of TCA cycle as it plays a central role in amino group metabolism. Kataria *et al.* (2010) also assessed the effect of extreme ambient temperature associated stress on fuelling of TCA cycle in hepatic cells of *Marwari* goat and reported that whenever a hepatocyte needs fuel for TCA cycle, the activity of enzyme glutamate dehydrogenase increases making alpha-ketoglutarate available for TCA cycle. Higher levels of serum ornithine carbamoyl transferase during hot ambience suggested higher activity of urea cycle as it is an important enzyme of urea cycle. Determination of its serum level is also important for detection of liver diseases and hepatocellular damage. Increased activity of arginase enzyme also suggested activation of urea cycle in heat stressed animals. It catalyses final step of urea cycle. Serum γGT is commonly used indicator of hepatobiliary disease in equines and ruminants (Saeed and Hussain, 2006).The determination of 5'-nucleotidase (5'-NT) has been most used in the cases where rise in serum ALP activity is there.

Glucose 6-phosphatase hydrolyzes glucose-6-phosphate resulting in the creation of a phosphate group and free glucose.Glucose is then exported from the cell via glucose transporter membrane proteins. This catalysis completes the final step in gluconeogenesis and glycogenolysis and therefore plays a key role in the homeostatic regulation of

blood glucose levels. Hot ambience probably served as a stressor and in order to maintain the blood glucose the activity of enzyme was higher. In present study the higher activity of serum ALD was related with higher utilisation of glucose through glycolysis to generate energy and also in the synthesis of glucose by gluconeogenesis in the stress period. Further higher ALD activity also helped in the generation of more glyceraldehydes –3-phosphate which can also be utilized for the synthesis of fat in the body (Kour and Kataria, 2011). Aldolase is one of the seven glycolytic enzymes, which function reversibly for gluconeogenesis, an important mechanism for generation of glucose in ruminants (Abdel-Fattah *et al*., 2002). Role of aldolase in carbohydrate metabolism explains its functional significance. Regulated glycolysis and gluconeogenesis prevent the futile cycling with accompanying loss of ATP and energy (Kour and Kataria, 2011).

The present study provided mean values of metabolic enzymes i.e. SDH, MDH, GD, G-6-PD, OCT, GGT, 5'NT, G-6-P, ARG, ALD etc. in serum of healthy donkeys of arid tract in India at one stand to find out ambience related metabolic changes. The variations observed in the present study could help in realistic assessment of the management practice, nutrition and diagnosis of disease conditions as all the enzymes investigated in this investigation belonged to the metabolism. The pattern of variation showed the activation of gluconeogenesis, hexose monophosphate shunt and urea cycle during hot ambience in donkeys of arid tract.

CONCLUSION

It was concluded that higher activity of metabolic regulators during hot ambience in otherwise healthy donkeys indicated the modulation of physiological mechanisms to support the metabolic pathways in adverse climatic conditions. Activation of gluconeogenesis probably helped the animals to combat heat stress. The present study provides reference values of some serum enzymes regulating key metabolic reactions at one stand for use in future research and to help in clinical cases for disease diagnosis.

REFERENCES

Abdel-Fattah, M., Moussa, S.Z., El-Hindy, H.M.,Gouda, E.M. (2002). Gluconeogenic behaviour of camel hepatocytes (Camelus dromedarius) by using (1-C14) propionate and (1-C14) lactate as substrates. *J Camel Prac Res*. **9 (2)**, 107-114.

Alemu, P., Forsyth, G.W., Searcy, G.P. (1977). A comparison of parameters used to assess liver damage in sheep treated with carbon tetrachloride. *Can J Comp Med*. **41**, 420- 427.

Brown, A.W., Grisolia, S. (1959). *J Lab Clin Med*. **54**, 417-420. (Cited from King, 1965).

Hanson, R.W., Ballard, F.J. (1967). Lipid Synthesis in Liver and Adipose Tissue from Ruminants *Biochem J*. **105**, 529-529.

Kataria, N., Kataria, A.K. (2010). Assessment of stress due to hot ambience in donkeys from arid tracts in India. *J Stress Physiol Biochem*. **6(4)**, 12-17.

Kataria, N., Kataria, A.K., Chaturvedi, M., Sharma, A. (2011).Changes in serum enzymes levels associated with liver functions in stressed *Marwari* goat. *J Stress Physiol Biochem*. **7(1)**, 13-19.

Kataria, N., Kataria, A. K. (2006). Endocrine and metabolic responses of Marwari sheep in arid tract. *Slov Vete Res*. **43(3)**, 135-142.

Kataria, N., Kataria, A.K., Joshi, A. (2010). Fuelling of TCA cycle in hepatic cells of *Marwari* goat during ambient temperature associated stress. *J Stress Physiol Biochem* **6(4)**, 5-11.

King, J. (1965). In: Practical clinical enzymology.

London: D.Van Nostrand Company Ltd., 1-301.

Kour, G., Kataria, N. (2011). Ambient temperature associated variations in serum arginase and aldolase in *Marwari* goat. *Vet Prac,* **12**, 124-127.

Manning, R.T., Grisolia, S. (1957). *Proc Soc exp Biol Med*. **95**, 225-226 (Cited from King, 1965).

Saeed, A., Hussain, M.M. (2006). Influence of age and sex on various serum enzyme activities of camels. *J Camel Prac Res*. **13(2)**, 149-155.

Sibley, J.A., Lehninger, A.L. (1949). Aldolase in serum and tissue of tumor bearing animals. *J Nat Cancer Inst*. **9**, 303-309.(Cited from King,1965).

Snedecor, G.W., Cochran, W.G. (1967). In: Statistical Methods. 6th ed. New Delhi:Oxford & IBH Publishing Co. 45-83.

Varley, H. (1988). Tests in liver and biliary tract disease. In: Practical Clinical Biochemistry. New Delhi: CBS publishers. 158-467.

Wolf, P.L., Williams, D. (1973). In: Practical Clinical Enzymology. New York: Wiley-Inetrscience Publication, John Wiley & Sons. 37- 85.