

## PHYSIOLOGICAL CONSEQUENCES VERSUS SCALING OF ENVIRONMENTAL STRESS IN MURRAH BUFFALOES FROM ARID TRACTS OF RAJASTHAN

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### ABSTRACT

Environmental phenomena in arid tracts can produce immense stress on animals and scaling of stress can help in evaluating physiological modulations. Scarcity of research on this aspect in buffaloes tended to initiate the present exploration to assess physiological consequences along with scaling of environmental stress in *Murrah* buffaloes from arid tracts of Rajasthan during extreme ambiances. Environmental factors were evaluated on the basis of temperature humidity index and heat load index values. Physiological consequences were assessed by measuring serum cortisol and serum glutathione reductase. Apparently healthy female buffaloes were screened during moderate, extreme hot, rainy and extreme cold ambiances. The overall mean values of serum cortisol and glutathione reductase were significantly ( $p \leq 0.05$ ) higher during rainy, extreme hot and extreme cold ambiances as compared to

respective moderate mean value. During rainy ambience, the per cent variation was found to be utmost for serum cortisol and glutathione reductase. It can be explained on the basis of serum cortisol that between heifers, pre-pubertals divulged higher effect of extreme ambiances. Amongst another group of female buffaloes, non-pregnant milch and primipara revealed greater thrust of extreme ambiances. Modulations in serum glutathione reductase revealed the existence of oxidative stress in all the buffaloes screened and the bang was greatest during rainy ambience. The essence of the present investigation was that rainy ambience produced the progression of greater scale of stress in the buffaloes with modulations in

the physiological reactions to a level which can institute upsetting of health of buffaloes. Environmental factors during extreme ambiances caused stress to the buffaloes of all physiological states. Physiological consequences were of utmost magnitude during rainy ambience. Rainy ambience divulged maximum values of heat load index and temperature humidity index exhibiting peak effect. Upshots of present study insinuated that the ration of buffaloes must be enriched with sufficient antioxidants in the form of vitamins and minerals. Marginal owners must be persuaded to monitor the health of buffaloes from arid tracts frequently.

**KEYWORDS:** Ambience, cold, cortisol, glutathione reductase, rainy.

## INTRODUCTION

Rajasthan is the part of hot Indian arid zone. Drastic variations in temperatures of the environment put stress on the native animals. Arid tracts of Rajasthan can be grouped into moderate, hot, rainy and extreme cold ambiances including the months of October-November; April-May-June; July-August-September and December-January, respectively with some intervening period (February-March months). Extreme ambient temperatures and humidities can modulate physiological consequences.<sup>[1,2,3,4,5]</sup> Serum cortisol is a good indicator of stress in the animals and the native animals to arid tract of Rajasthan living under natural husbandry conditions are repeatedly beleaguered owing to cruel ambience ensuing in gigantic economical pasting to marginal animal owners. Researchers from time to time have tried hard to assess stress by measuring cortisol levels.<sup>[6,7,8,9,10,11]</sup> However, there is paucity of research on this aspect in buffaloes especially from arid tracts of Rajasthan. Glutathione reductase is one of the biomarkers of oxidative stress. There are many reasons of oxidative stress starting from abiotic to biotic stressors.<sup>[12,13,14,15,16,17,18,19]</sup> Environmental stressors can be evaluated on the basis of temperature humidity index and heat load index.<sup>[20,21,22,23]</sup> Despite of the fact that buffaloes are important economical scaffolding of poor and marginal animal owners, very little scientific consideration has been offered. In the arid tracts of Rajasthan, these animals face many stressors, however, environmental factors are chief stressors to these animals. Looking towards the importance of buffaloes and paucity of research on this aspect, present investigation was launched to evaluate physiological consequences due to environmental stress in *Murrah* buffaloes from arid tracts of Rajasthan. The data produced in the investigation will help as building blocks for future research.

## MATERIALS AND METHODS

Apparently healthy *Murrah* female buffaloes (800) incorporating buffalo heifers and buffaloes in different physiological states according to milch and parity status ageing one year old to 8 years of age were screened from private dairies located in and around Bikaner district, Rajasthan, India under the permission of Institutional Animal Ethics Committee, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner, Rajasthan. Collection of blood samples was made to harvest serum during moderate (October-November), extreme hot (April-May-June), rainy (July-August-September) and extreme cold (December-January) ambiances. Category of buffalo heifers included pre-pubertal and post-pubertal, each encompassing 40 buffaloes. Buffaloes were also grouped as A and B. Sub-group A comprised of non-pregnant milch (40); pregnant milch (40) and pregnant dry (40) buffaloes. Sub-group B incorporated primipara (60) and multipara (60) buffaloes. Serum cortisol was measured by chemiluminescence assay. Serum glutathione reductase was determined by standard method<sup>[24]</sup> with little modifications. Temperature humidity index values was computed as per standard equation.<sup>[25]</sup> Heat load index was calculated as per standard protocol.<sup>[26]</sup> Data were expressed as mean $\pm$ SE of mean and computer programmes were employed to compute means and standard error (<http://www.miniwebtool.com>). The importance of the impacts was also assessed.<sup>[27]</sup>

## RESULTS AND DISCUSSION

Environmental factors were assessed by obtaining mean  $\pm$  SEM values of temperature humidity index and heat load index by collecting data of environmental elements from Bikaner district, Rajasthan during varying ambiances. The mean  $\pm$  SEM values of temperature humidity index and heat load index are presented in table 1. The mean value of each obtained during moderate ambience was considered as control value.

**Table 1: Mean  $\pm$  SEM values of environmental variables (EV<sub>s</sub>) of Bikaner district, Rajasthan during varying ambiances of sampling period (N=40).**

S.no.	Environmental Variables	Mean $\pm$ SEM values during varying ambiances			
		Moderate	Extreme Hot	Rainy	Extreme Cold
1	THI <sub>MinET</sub>	65.29 <sup>b</sup> $\pm$ 0.12 (53.71-74.74)	80.29 <sup>b</sup> $\pm$ 0.12 (67.70-88.10)	82.38 <sup>b</sup> $\pm$ 0.14 (73.01-90.00)	50.37 <sup>b</sup> $\pm$ 0.13 (40.00-68.08)
2	THI <sub>MaxET</sub>	76.49 <sup>b</sup> $\pm$ 0.26 (70.11-78.84)	87.29 <sup>bd</sup> $\pm$ 0.26 (79.00-92.00)	89.39 <sup>b</sup> $\pm$ 0.11 (82.10-96.10)	63.39 <sup>b</sup> $\pm$ 0.20 (46.00-71.00)
3	THI <sub>AET</sub>	71.38 <sup>bc</sup> $\pm$ 0.11 (60.22-77.31)	85.48 <sup>b</sup> $\pm$ 0.11 (72.00-92.4)	86.23 <sup>b</sup> $\pm$ 0.12 (82.20-93.08)	62.58 <sup>b</sup> $\pm$ 0.16 (50.00-69.08)
4	HLI <sub>MaxET</sub>	76.31 <sup>b</sup> $\pm$ 0.40 (61.01-89.91)	81.13 <sup>b</sup> $\pm$ 0.30 (64.04-87.01)	92.48 <sup>b</sup> $\pm$ 0.37 (70.16-96.19)	52.18 <sup>b</sup> $\pm$ 0.48 (34.20-75.09)
5	HLI <sub>MinET</sub>	65.05 <sup>b</sup> $\pm$ 0.09 (54.01-78.91)	67.22 <sup>b</sup> $\pm$ 0.09 (58.42-73.92)	70.86 <sup>b</sup> $\pm$ 0.14 (60.06-83.94)	34.00 <sup>b</sup> $\pm$ 0.13 (22.44-43.78)
6	HLI <sub>AET</sub>	70.07 <sup>b</sup> $\pm$ 0.60 (61.01-83.02)	76.05 <sup>b</sup> $\pm$ 0.36 (63.39-79.61)	83.06 <sup>b</sup> $\pm$ 0.36 (63.10-89.16)	46.00 <sup>b</sup> $\pm$ 0.33 (30.00-61.95)

N = Number of observations

THI = Temperature humidity index

AET = Average Environmental temperature

MaxET = Maximum Environmental temperature

MinET = Minimum Environmental temperature

HLI = Heat load index

'<sup>b</sup>' marks significant ( $p \leq 0.05$ ) differences among mean values for a row.

Figures in the parentheses are ranges.

The variables of temperature humidity index were minimum, maximum and average. They were grouped according to inclusion of minimum, maximum and average required environmental elements for the calculation. The values among ambiances varied significantly ( $p \leq 0.05$ ). Rainy ambience exhibited maximum values of all the three elements of temperature humidity index as compared to respective values during moderate, extreme hot and extreme cold ambiances.

The elements of heat load index were minimum, maximum and average. The values among ambiances varied significantly ( $p \leq 0.05$ ). Rainy ambience showed maximum values of all the three elements of HLI as compared to respective values during moderate, extreme hot and extreme cold ambiances. In the arid tract of Rajasthan, April to September months generally encounter higher ambient temperature. Workers have revealed bang of higher and lower ambient temperatures on different physiological consequences of the animals.<sup>[26,28,29,30,31]</sup> Utmost temperature humidity index of moderate period revealed vast range, however,

previous workers have reported more or less similar temperature humidity index values during moderate ambience from Bikaner, Rajasthan as observed in present investigation.<sup>[32,33,34]</sup> Forbearance of the animals to temperature humidity index shows their physiological adjustment.

The heat load index can be established as a parameter of the environmental heat load which is transferred to buffaloes and is a sign of physiological stress to the animals. Heat load index in addition to temperature humidity index can be considered as an important environmental index affecting physiology of animals.<sup>[34,35,36]</sup> The mean value of maximum heat load index in present study corroborated the earlier finding from the Bikaner district, Rajasthan.<sup>[32]</sup> Prolonged exposure of buffaloes to extremely high environmental temperatures and high relative humidity can reduce heat loss ability. Gratuitous heat load can give rise to considerable production loss. Extended increase in body temperature can cause tissue and organ damage. It has become important to reorganize the critical levels of various environmental elements related to combine the bang of environment on the thermal comfort of buffaloes.

It can be restated that moderate months of the Bikaner, Rajasthan on an average lived through a higher range of temperature humidity index in terms of minimum, average and maximum, respectively. Per cent augmentation in the respective values of temperature humidity index as minimum, maximum and average was found to be maximum during rainy ambience followed by extreme hot as compared to respective moderate ambience mean value.

Looking towards the vast geographical and environmental characteristics of India, it is compulsory to set up soothe zone of each area having diversity. It has been observed that buffaloes native to these areas are adjusted with their separate thermal zones, however, it is important to note that adjustments should not be on the cost of production and health. It is also not prudent to extrapolate ranges of environmental temperatures and humidities which are pertinent to other animals of different geographical areas on buffaloes as different animal species have changeable level of responses to temperature and relative humidity, therefore, the heat stress calculation is unique for a particular species.<sup>[20]</sup>

### Serum cortisol

Mean  $\pm$  SEM values of serum cortisol are presented in table 2. The moderate overall mean value of serum cortisol was more or less in agreement to the values recorded by previous workers in native animals from arid tracts.<sup>[6,9,10,14,26,37,38]</sup>

**Table 2: Mean  $\pm$  SEM values of serum cortisol (nmolL<sup>-1</sup>) in the female *Murrah* buffaloes during varying ambiances.**

S. No.	Effects	Mean $\pm$ SEM values during ambiances			
		Moderate	Extreme hot	Rainy	Extreme cold
1.	Ambience Overall values (200)	34.08 <sup>b</sup> $\pm$ 2.90	73.005 <sup>b</sup> $\pm$ 1.80	93.78 <sup>b</sup> $\pm$ 3.52	42.83 <sup>b</sup> $\pm$ 1.62
2.	Age group categorization (I & II categories)				
I.	<b><i>Murrah</i> Heifers, 1-3.5 years (80), categorization as a &amp; b</b>				
	Overall values (80)	31.18 <sup>bg</sup> $\pm$ 1.07	71.20 <sup>bg</sup> $\pm$ 1.10	90.26 <sup>bg</sup> $\pm$ 1.11	41.21 <sup>bg</sup> $\pm$ 1.20
A	Pre-pubertal (40)	30.11 <sup>bd</sup> $\pm$ 0.05	70.10 <sup>bd</sup> $\pm$ 0.04	89.15 <sup>bd</sup> $\pm$ 0.05	40.01 <sup>bd</sup> $\pm$ 0.05
B	Post-pubertal (40)	32.25 <sup>bd</sup> $\pm$ 0.04	72.30 <sup>bd</sup> $\pm$ 0.04	91.37 <sup>bd</sup> $\pm$ 0.05	42.42 <sup>bd</sup> $\pm$ 0.04
II.	<b>Female <i>Murrah</i> buffaloes, 3.5-8 years (120), categorization as group A &amp; B</b>				
	Overall values (120)	36.99 <sup>bg</sup> $\pm$ 2.22	74.81 <sup>bg</sup> $\pm$ 1.35	97.31 <sup>bg</sup> $\pm$ 2.41	44.46 <sup>bg</sup> $\pm$ 1.59
	<b>Group A (120), Physiological states: Pregnancy and milch status</b>				
A	Non-pregnant milch (40)	33.015 <sup>be</sup> $\pm$ 0.945	72.84 <sup>be</sup> $\pm$ 0.74	93.20 <sup>be</sup> $\pm$ 1.10	41.64 <sup>be</sup> $\pm$ 0.74
B	Pregnant milch (40)	37.28 <sup>be</sup> $\pm$ 0.96	74.20 <sup>be</sup> $\pm$ 0.905	97.16 <sup>be</sup> $\pm$ 1.05	44.58 <sup>be</sup> $\pm$ 1.08
C	Pregnant dry (40)	40.70 <sup>be</sup> $\pm$ 0.815	77.40 <sup>be</sup> $\pm$ 1.30	101.57 <sup>be</sup> $\pm$ 0.86	47.16 <sup>be</sup> $\pm$ 1.06
	<b>Group B (120), Physiological states: Parity</b>				
A	Primipara (60)	36.12 <sup>bf</sup> $\pm$ 2.23	73.83 <sup>bf</sup> $\pm$ 1.18	96.30 <sup>bf</sup> $\pm$ 2.48	43.50 <sup>bf</sup> $\pm$ 1.50
B	Multipara (60)	37.93 <sup>bf</sup> $\pm$ 2.16	75.49 <sup>bf</sup> $\pm$ 1.61	98.31 <sup>bf</sup> $\pm$ 2.34	45.42 <sup>bf</sup> $\pm$ 1.69

- i. Figures in the parenthesis = Number of *Murrah* buffaloes
- ii. 'b' = Significant ( $p \leq 0.05$ ) differences among mean values for a row.
- iii. 'd' = Significant ( $p \leq 0.05$ ) differences between mean values of heifers for an ambience
- iv. 'e' = Significant ( $p \leq 0.05$ ) differences among mean values of Group A for an ambience
- v. 'f' = Significant ( $p \leq 0.05$ ) differences between mean values of Group B for an ambience
- vi. 'g' = Significant ( $p \leq 0.05$ ) differences among overall values of heifers and buffaloes for an ambience.

### Variations in values of serum cortisol during different ambiances

Mean values of serum cortisol were significantly ( $p \leq 0.05$ ) higher during extreme hot, rainy and extreme cold ambiances as compared to moderate mean value. The per cent variation in the value of serum cortisol was observed to be utmost during rainy ambience. Previous

workers are of the opinion that cortisol level may rise owing to different stress factors and a doubling of serum cortisol can be instituted as the increase required for the occurrence of stress.<sup>[9,26]</sup> In the present investigation, increase was higher during rainy ambience than extreme hot ambience. Earlier researchers have also associated increase in the serum cortisol levels with hot ambience in animals from arid tracts.<sup>[6,9,10,26,37]</sup> Cortisol lifts glucose levels by the glucogenolytic and gluconeogenetic pathways. Increase in cortisol is the physiological consequence of stress. Heat stress can cause a stumble of alterations in the physiological mechanisms to fight the stress. Higher cortisol level substantiated the significance in meeting the energy upheaval during the stress period.<sup>[6]</sup> Cortisol enhances the mechanism of an animal in response to stress in terms of psychobiological resilience to oxidative damage which depends upon the period of exposure.<sup>[6,9,26]</sup>

The findings of present investigation regarding cortisol exhibited its function as one of the causative factors in the progression of oxidative stress. Upshots of present exploration showed that impact of rainy ambience which was maximum in terms of cortisol lifts followed by extreme hot and cold ambiences. It can be reiterated that cortisol system was maximally modulated during rainy ambience as compared to extreme hot and cold ambiences.

#### **Effect of physiological states of buffaloes on serum cortisol**

Significant ( $p \leq 0.05$ ) changes among all the groups of buffaloes were observed in each ambience. Effect of rainy ambience was maximum on all the buffaloes. In heifers, post-pubertal had significantly ( $p \leq 0.05$ ) higher values of serum cortisol in each ambience as compared to pre-pubertal. In both the types, maximum mean values were observed in rainy ambience as compared to moderate ambience followed by extreme hot and extreme cold ambiences. Maximum per cent variation was revealed by pre-pubertal buffaloes.

Among group A buffaloes, pregnant-dry had significantly ( $p \leq 0.05$ ) higher values of serum cortisol in each ambience as compared to others. In all the three types, maximum mean values were observed in rainy ambience as compared to moderate ambience followed by extreme hot and extreme cold ambiences. In comparative terms, all the types divulged higher per cent changes during rainy ambience. Maximum per cent changes were revealed by non-pregnant milch as compared to pregnant milch and pregnant dry buffaloes.

In group B animals, multipara buffaloes had significantly ( $p \leq 0.05$ ) raised values of serum cortisol in ambience as compared to primipara buffaloes. In both the types, maximum mean

values were found in rainy ambience as compared to moderate ambience followed by extreme hot and extreme cold ambiances. Primipara buffaloes revealed greater per cent variations in extreme hot, rainy and extreme cold ambiances as compared to multipara buffaloes. Alterations in serum cortisol due to physiological states have been reported [26]. Increased values of serum cortisol could be ascribed to stress. Higher cortisol is essential for energy homeostasis owing to its metabolic role as an energy modulator through gluconeogenesis and lipolysis. [26,28]

### Serum glutathione reductase

Mean  $\pm$  SEM values of serum glutathione reductase are presented in table 3. The overall mean value of moderate period was close to the values recorded by researchers in native animals from arid tracts. [12,13,14,15,26] Serum glutathione reductase is an important biomarker of oxidative stress.

**Table 3: Mean  $\pm$  SEM values of serum glutathione reductase (kU L<sup>-1</sup>) in the female *Murrah buffaloes* during varying ambiances.**

S. No.	Effects	Mean $\pm$ SEM values during ambiances			
		Moderate	Extreme hot	Rainy	Extreme cold
1.	Ambience Overall values (200)	13.27 <sup>b</sup> $\pm$ 0.25	39.51 <sup>b</sup> $\pm$ 5.49	71.435 <sup>b</sup> $\pm$ 3.415	20.26 <sup>b</sup> $\pm$ 0.745
2.	Age group categorization (I & II categories)				
I.	<i>Murrah</i> Heifers, 1-3.5 years (80), categorization as a & b				
	Overall values (80)	13.02 <sup>bg</sup> $\pm$ 3.99	34.02 <sup>bg</sup> $\pm$ 2.01	68.02 <sup>bg</sup> $\pm$ 3.0	19.52 <sup>bg</sup> $\pm$ 4.49
A	Pre-pubertal (40)	9.03 <sup>bd</sup> $\pm$ 0.003	32.01 <sup>bd</sup> $\pm$ 0.003	65.02 <sup>bd</sup> $\pm$ 0.002	15.03 <sup>bd</sup> $\pm$ 0.004
B	Post-pubertal (40)	17.01 <sup>bd</sup> $\pm$ 0.001	36.03 <sup>bd</sup> $\pm$ 0.004	71.02 <sup>bd</sup> $\pm$ 0.003	24.02 <sup>bd</sup> $\pm$ 0.003
II.	Female <i>Murrah</i> buffaloes, 3.5-8 years (120), categorization as group A & B				
	Overall values (120)	13.52 <sup>bg</sup> $\pm$ 1.160	45.01 <sup>bg</sup> $\pm$ 2.309	74.85 <sup>bg</sup> $\pm$ 2.446	21.01 <sup>bg</sup> $\pm$ 2.306
	Group A (120), Physiological states: Pregnancy and milch status				
A	Non-pregnant milch (40)	11.51 <sup>be</sup> $\pm$ 0.080	41.012 <sup>be</sup> $\pm$ .159	70.53 <sup>be</sup> $\pm$ 0.239	17.02 <sup>be</sup> $\pm$ 0.161
B	Pregnant milch (40)	13.52 <sup>be</sup> $\pm$ 0.079	45.015 <sup>be</sup> $\pm$ .158	75.02 <sup>be</sup> $\pm$ 0.159	21.012 <sup>be</sup> $\pm$ 0.161
C	Pregnant dry (40)	15.53 <sup>be</sup> $\pm$ 0.079	49.015 <sup>be</sup> $\pm$ .161	79.007 <sup>be</sup> $\pm$ 0.159	25.012 <sup>be</sup> $\pm$ 0.159
	Group B (120), Physiological states: Parity				
A	Primipara (60)	13.025 <sup>bf</sup> $\pm$ 0.213	44.015 <sup>bf</sup> $\pm$ 0.424	73.69 <sup>bf</sup> $\pm$ 0.477	20.01 <sup>bf</sup> $\pm$ 0.425
B	Multipara (60)	14.02 <sup>bf</sup> $\pm$ 0.213	46.01 <sup>bf</sup> $\pm$ 0.425	76.01 <sup>bf</sup> $\pm$ 0.424	22.02 <sup>bf</sup> $\pm$ 0.212

- i. Figures in the parenthesis = Number of *Murrah* buffaloes
- ii. 'b' = Significant ( $p \leq 0.05$ ) differences among mean values for a row.
- iii. 'd' = Significant ( $p \leq 0.05$ ) differences between mean values of heifers for an ambience
- iv. 'e' = Significant ( $p \leq 0.05$ ) differences among mean values of Group A for an ambience

- v. 'f' = Significant ( $p \leq 0.05$ ) differences between mean values of Group B for an ambience
- vi. 'g' = Significant ( $p \leq 0.05$ ) differences among overall values of heifers and buffaloes for an ambience

### **Variations in values of serum glutathione reductase during varying ambiences**

The moderate mean values of serum glutathione reductase were significantly ( $p \leq 0.05$ ) higher during extreme cold, hot and rainy ambiences as compared to moderate mean value. The per cent variation in the value of serum glutathione reductase was found to be maximum during rainy ambience. Among extreme ambiences, the overall mean value of rainy ambience was maximum and that of extreme cold was minimum.

Earlier workers have related extreme ambient temperatures with the higher generation of free radicals leading to production of oxidative stress.<sup>[12,13,14,15,26]</sup> Elevated levels of serum glutathione reductase in present investigation divulged the bang of extreme ambient temperatures. It is to mention that higher temperature period was observed to be related with higher production of free radicals since values of glutathione reductase were found to be utmost. Serum glutathione reductase is an important part of antioxidant defense system and is a powerful endogenous free radical forager.

### **Effect of physiological states on serum glutathione reductase values**

Female buffaloes were divided into two major groups (heifers and milch and parity states) in all the four ambiences. The moderate mean value in each case was taken as control. The alterations due to extreme ambiences were significant ( $p \leq 0.05$ ).

Between heifers, post-pubertal had significantly ( $p \leq 0.05$ ) haughty values of serum glutathione reductase in each ambience as compared to pre-pubertal. In both the types of buffalo heifers, higher mean values were observed in rainy ambience as compared to moderate ambience and lowest values in extreme cold ambience. Simultaneously, higher per cent variations were observed during rainy ambience. Post-pubertal heifers divulged greater per cent variation in each ambient temperature period. In group A buffaloes, pregnant- dry had significantly ( $p \leq 0.05$ ) higher values of serum glutathione reductase in each ambience. In all the three types, utmost mean values were found in rainy and lowest values in extreme cold as compared to moderate ambience. Maximum per cent change was depicted by non-pregnant milch during all ambient temperature periods. In group B buffaloes, multipara

showed significantly ( $p \leq 0.05$ ) higher values of serum glutathione reductase in each ambient temperature period as compared to primipara buffaloes. In both the types, maximum mean values were observed in rainy as compared to moderate ambient temperature period. Multipara tended to divulge higher per cent changes in each ambient temperature period. Changes in serum glutathione reductase owing to physiological states have been reported by earlier workers.<sup>[26]</sup>

In the present study, scaling of environmental stress was done on the basis of determination of intensity of environmental factors by measuring temperature humidity index and heat load index. Physiological consequences were assessed on the basis of serum cortisol and glutathione reductase. Rainy ambience appeared to be the most stressful ambient period for all the buffaloes screened.

## CONCLUSION

It can be concluded that environmental stress was produced in female *Murrah* buffaloes during extreme ambiances. Impact was maximum during rainy ambience which was based on higher temperature humidity index and heat load index values. Serum cortisol and glutathione reductase showed alterations during rainy, extreme hot and extreme cold ambiances. Maximum changes were found during rainy ambience.

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