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Alterations of hemato-biochemical responses on adaptability of buffaloes in eastern plane zone of Uttar Pradesh

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Abstract

In tropical climate like India an environmental temperature above the temperature of thermo-neutral zone causes more heat gain than the heat lost in the animal body and animal suffer from thermal stress. Study was carried out to explore the variation in Hemato-biochemical responses using various techniques on adaptability of buffaloes in eastern plane zone of Uttar Pradesh. During summer season, there was significant (p<0.01) reduction in plasma glucose level (mg/dl), Blood Urea Nitrogen level (mg/dl), plasma total protein level (g/dl), in comparison to spring season in buffaloes but there was significant (p<0.01) increase in plasma cortisol level in summer in comparison to spring season in all the three districts of eastern plane zone of Uttar Pradesh. Haemoglobin (g/dl) concentration, Packed cell volume, Total erythrocyte count was significantly (p<0.01) lower in summer season in comparison to spring season in buffaloes but Total leucocyte count was significantly (p<0.01) higher in summer season in comparison to spring season in all the three districts. In conclusion, heat stress has unfavorable effect on adaptability and physio-biochemical responses of buffalo.

Keywords: Buffalo, heat stress, hemato-biochemical parameter, thermal adaptability

Introduction

Animals automatically react to stress in unfavourable environments, which can have negative consequences ranging from discomfort to death. Across the world, particularly in tropical and temperate regions, climate change is one of the biggest threats to the survival of many animal species, ecosystems, and livestock production systems. The normal body temperature of dairy animal ranges between 38.4- 39.1 °C in the thermo-neutral zone from 16 °C to 25 °C as per Yousef (1985)^[21]. Consequently, animals experience thermal stress when the outside temperature rises above the thermoneutral zone in a tropical country like India. This is because animals gain more heat than they lose from their bodies. Owing to increases in body surface temperature, heart rate, respiration rate (RR), and rectal temperature (RT), which have an impact on farm animals' ability to reproduce, consume feed, and produce. According to Kadokawa et al. (2012)^[8], animals experiencing heat stress high enough to impact milk supply and fertility were shown to have RT >39.0 °C and RR >60/min. Thus, species, breed, and productivity can all have an impact on thermal stress in homeothermic animals. Previous research has indicated that native breeds survive better than non-native breeds and exhibit superior performance. It is possible that this is because non-native genes are not able to express or adapt to tropical environments, leading to the observed differences in native breeds' survival and performance.

Preventing heat stress-related economic loss and enhancing the welfare of buffaloes can be achieved through a deeper comprehension of the physiological changes that occur in these animals under harsh environmental circumstances. Thus, the purpose of this study was to investigate the differences in hemato-biochemical reactions regarding the adaptation of buffaloes in Uttar Pradesh's eastern plane zone.

Materials and Methods Study Site

Department of Veterinary Physiology and Biochemistry, ANDUAT, Kumarganj, Ayodhya, Uttar Pradesh was the site of the study. The institutional animal ethics committee has accepted the experiment's design. The studies were conducted in the Eastern Plane Zone of Uttar Pradesh in the districts of Ayodhya, Mau, and Bhadohi during the spring (March) and summer (June) of 2021. In spring (March) and summer (June), all the climate variables were recorded twice a day from 7:30 am to 2:30 pm.

Management and design: The investigation involved the random selection of eighteen nursing Murrah breed buffaloes. The buffaloes included in the study ranged in age from 6 to 10 years and weight from 450 to 600 kg. The animals were split up into three distinct categories, with one group of six animals in each district. For the trial, nursing animals from field environments or dairy farms were employed. The animals were housed in an open space with an asbestos roof and a brick floor. The animals were allowed to roam freely throughout the house in order to get to the water and food supplies. Water and food were available to the animals at all times. Mustard cake, maize, wheat bran, rice bran, mineral mixture, and salt made up the concentrated mixture.

Collection of Blood samples and observation: A jugular vein puncture was used to obtain a blood sample (10 ml/buffalo) in the afternoon between 1:30 and 2:30 pm in sterile tubes containing EDTA with the least amount of disturbance to the animal. In the spring and summer, blood samples from the same experimental animals were taken once a week. The "Bovine Cortisol ELISA kit" (Catalogue No.E0110Bo) from Bioassay Technology Laboratory, 501 Changsheng S Rd, Nanhu Dist, Jiaxing, Zhejiang, China, was used for plasma collection and hemo-biochemical analysis, including plasma glucose, total plasma protein, blood urea nitrogen (BUN), cortisol hormone, etc.

Recording of Hematological Parameters

The haematological parameters like Haemoglobin, Total erythrocyte count (TEC), Total leukocyte count (TLC) and Packed cell volume (PCV) were recorded.

Estimation of Haemoglobin

Haemoglobin (Hb) was estimated in gm % by using method as described by Schalm *et al.* (1975) ^[16].

Total Erythrocyte Count (TEC) and Total Leucocyte Count (TLC)

Total Erythrocyte Count (TEC) and Total Leucocyte Count (TLC) were enumerated by haemocytometer method as described by Jain (1986) ^[5]. TEC was expressed in millions per cubic millimeter (10^{6} /cmm) and TLC expressed in thousand per cubic millimeter (10^{3} /cmm).

Estimation of Packed Cell Volume (PCV)

Packed Cell Volume was determined by using Wintrobe haematocrit tube as per the standard method described by Jain (1986)^[5] and expressed in terms of percentage (%).

Estimation of Glucose

Glucose was estimated in plasma samples of buffalo by using GOD-POD kits from Span Diagnostics Ltd. The principle of the assay (Kaplan, 1984) ^[7] is glucose oxidase (GOD) oxidizes glucose to gluconic acid and hydrogen peroxide. In presence of enzyme peroxidase, released hydrogen peroxide is coupled with phenol and 4-aminoantipyrine (4-AAP) to form coloured quinoneimine dye. Absorbance of coloured dye is measured at 505 nm and is directly proportional to glucose concentration in the sample. Glucose $+ O_2 + H_2O$

Glucose Oxidase \rightarrow Gluconic Acid +H₂O₂

 H_2O_2 + Phenol + 4-AAP Peroxidase \rightarrow Quinoneimine dye + H_2O

Calculation

Plasma glucose (mg/dl) =
$$\frac{\text{Absorbance of Test}}{\text{Absorbance of Standard}} X 100$$

Glucose Concentration in mmol / L= Glucose Concentration in mg /dL X 0.05551

Urea Analysis

Blood Urea was measured in buffalo plasma samples using the DAM Assay kit from Arkray Healthcare Pvt. Ltd. in Japan (MBK Urea kits). The basic idea behind the test (Roberts *et al.*, 2008) ^[13] is that urea, in the presence of thiosemicarbazide, combines with hot, acidic dialcetylmonoxime to form a rose-purple coloured complex that can be detected colorimetrically. The sample did not need to be deproteinized.

Calculation of Results

Serum / plasma: Urea in mg/100 mL, (A) = $\frac{\text{O.D. of Test}}{\text{O.D of Std}} \times 30$

Urea nitrogen in mg/100 mL = (A) \times 0.467

Assay for Total Protein

The amount of total protein in buffalo plasma samples was measured using Autospan kits from Japan's Arkray Healthcare Pvt. Ltd. Peptide bonds in proteins react with cupric ions in an alkaline solution to generate a coloured chelate, the absorbance of which is evaluated at 578 nm (550-580 nm). This is the basic idea of the assay (Roberts *et al.*, 2008) ^[13]. The amount of Total Protein present in the sample is directly correlated with the final color's absorbance.

Protein + $Cu^{++} \rightarrow Alkaline pH Cu$ - Protein complex

Calculation of Results

Total Protein concentration $(g/dL) = \frac{\text{Absorbance of test}}{\text{Absorbance of standard}} \times 6.5$

Globulins = Total Protein - Albumin

Total Protein concentration in g/L = Total Protein concentration in g / dL $\times 10$

Result and Discussion Plasma Glucose

Tables 01, 02, and 03 show the results of the plasma glucose level (mg/dl) in buffaloes over the spring and summer. In Ayodhya, Bhadohi, and Mau districts, the mean plasma glucose level (mg/dl) of buffaloes during the spring season was 74.41±0.36, 74.47±0.38, and 74.41±0.37 mg/dl, respectively. The average plasma glucose levels in the buffaloes of the Ayodhya, Bhadohi, and Mau districts over the summer were 72.10±0.26, 72.11±0.26, and 72.00±0.26 mg/dl, respectively. The summer season in all three districts of Uttar Pradesh's eastern plane zone showed a significant (p < 0.01) drop in plasma glucose levels compared to the spring. According to Shafferi et al. (1981)^[17], an increase in respiration rate in a hot climate may be the cause of the drop in plasma glucose since it leads to the respiratory muscle utilising glucose more often. Similar findings of buffaloes' summertime plasma glucose levels were reported by Ronchi *et al.* (1995) ^[14] and Verma *et al.* (2000) ^[19].

Blood Urea Nitrogen (BUN)

The results of the buffaloes' spring and summer blood urea nitrogen levels (mg/dl) were displayed in Tables 01, 02, and 3. In the spring, the average blood urea nitrogen (mg/dl) in buffaloes was 16.93±0.22, 16.47±0.25, and 16.57±0.18 mg/dl in the districts of Ayodhya, Bhadohi, and Mau, respectively. In buffaloes from the districts of Ayodhya, Bhadohi, and Mau, the mean blood urea nitrogen (mg/dl) throughout the summer was 14.68±0.18, 14.72±0.18, and 14.62±0.16 mg/dl, respectively. The findings indicate that in all three districts of Uttar Pradesh's eastern plane zone, there was a substantial (p < 0.01) drop in blood urea nitrogen levels throughout the summer compared to the spring. Blood urea nitrogen levels are decreasing, and this can be explained by more urea nitrogen being reabsorbed from the blood into the rumen to make up for the lower concentration of ammonianitrogen caused by decreased feed consumption and digestible nitrogen use (Yousef, 1990)^[22]. Similar findings of a decrease in blood urea nitrogen levels throughout the summer have been documented in buffaloes by Ronchi et al. (1995)^[14], in Verma *et al.* (2000)^[19], and in Friesian calves by Kamal et al. (1989)^[6].

Plasma Total Protein

Tables 04,05,06 show the results of the plasma total protein level (g/dl) in buffaloes over the spring and summer. In the spring, the average plasma total protein level (g/dl) in buffaloes was 7.57±0.09, 7.55±0.10, and 7.65±0.08 g/dl in the districts of Ayodhya, Bhadohi, and Mau, respectively. In the summer, the average total protein levels in buffaloes from the districts of Ayodhya, Bhadowhi, and Mau were 6.58±0.10, 6.60±0.08, and 6.62±0.11 g/dl, respectively. The summer season in all three districts of Uttar Pradesh's eastern plane zone showed a substantial (p < 0.01) decrease in plasma total protein levels compared to the spring season. The decrease in dietary protein consumption during heat stress caused by higher THI levels in the summer may be the cause of the drop in plasma total protein level. The total plasma protein synthesis is markedly reduced in prolong dietary protein deficiency (William, 2005) [20]. Similar observation of reduction in total plasma protein level during heat stress is reported by Habeeb et al., (2007)^[4] in buffalo calves and Verma et al., (2000) [19] in murrah buffaloes. Rasooli et al., (2004)^[12] reported in hot summer significant increase in total plasma protein in Holstein heifer but there finding is not agreement with our finding in buffalo during summer season.

Plasma Cortisol

The concentration of plasma cortisol (ng/ml) has been presented in Table 04, 05, 06. In spring the mean plasma cortisol of buffalo was 4.67±0.21, 4.38±0.16 and 5.35±0.27 ng/ml in Ayodhya, Bhadohi and Mau districts respectively. The mean of plasma cortisol level during summer was 12.19±0.21, 11.84±0.34 and 12.09±0.22 ng/ml in buffaloes of Ayodhya, Bhadohi and Mau districts respectively. Results shows that in summer season there was significant (p < 0.01) increase in plasma cortisol level in comparison to spring season in all the three districts of eastern plane zone of Uttar Pradesh. The reason of increase in plasma cortisol level may be attributed due to in stressful condition Hypothalmopituitary adrenal axis is activated and increase in plasma cortisol level. Plasma cortisol helps in physiological adjustments of animal during stressful condition (Christison and Johnson, 1972) ^[2]. Similar observation of increase in plasma cortisol level during stressful condition was reported by Lakhani *et al.*, (2018)^[9] in murrah buffalo and Liu et al., (2020) ^[10] in Nili-Ravi buffalo Chaudhary et al., (2015)^[1] in Surti buffalo.

Haematological Parameters

Haemoglobin

Tables 07, 08, and 09 show the buffalo's hemoglobin concentration (g/dl) findings for the spring and summer seasons. The districts of Ayodhya, Bhadohi, and Mau had the mean \pm standard error of hemoglobin concentration during the spring, which was 11.45±0.03, 11.43±0.04, and 11.47±0.03 g/dl, respectively, for buffaloes. In the summer, the mean haemoglobin concentration in the districts of Ayodhya, Bhadohi, and Mau was 10.37±0.06, 10.35±0.06, and 10.30±0.05 g/dl, respectively. In all three districts, the summertime haemoglobin (g/dl) concentration was considerably (p < 0.01) lower than the springtime value. Erythropoiesis depression may be the cause of the summertime drop in hemoglobin concentration (Shebaita and Kamal, 1973)^[18]. Our results are in agreement with those of Yousef (1990)^[22], Daader *et al.* (1989)^[3], and Marai et al. (1995)^[11].

Packed Cell Volume (PCV)

Tables 07, 08, and 09 show the packed cell volume (%) of buffaloes during the spring and summer seasons. Ayodhya, Bhadohi, and Mau districts had the following mean \pm SE of packed cell volume (%) during the spring: 34.38 \pm 0.06, 34.49 \pm 0.05, and 34.30 \pm 0.06 consequently. In the districts of Ayodhya, Bhadohi, and Mau, the mean packed cell volume (%) throughout the summer was 33.28 \pm 0.03, 33.13 \pm 0.05, and 32.78 \pm 0.11%, respectively. In all three districts, the summer season had a considerably (p<0.01) lower packed cell volume than the spring season. Summertime decreases in packed cell volume (%) may be related to erythrocyte breakdown under conditions of temperature stress (Shebaita and Kamal, 1973) ^[18]. Our results are in agreement with those of Marai *et al.* (1995) ^[11].

Total Erythrocyte Count (TEC)

Table 10, 11, 12 displays the buffalo's total erythrocyte count (million/ μ l) for the spring and summer seasons. In buffaloes, the mean ± standard error of the total erythrocyte count throughout the spring was 6.71±0.02, 6.71±0.02, and 6.70±0.02 million/ μ l in the districts of Ayodhya, Bhadohi, and Mau, respectively. The mean temperature throughout

the summer was 5.76 ± 0.03 , 5.76 ± 0.03 , and 5.73 ± 0.03 million/µl in the districts of Ayodhya, Bhadohi, and Mau, in that order. In all three districts, the total erythrocyte count was considerably (*p*<0.01) lower in the summer than in the spring. According to Shafferi *et al.* (1981) ^[17], the breakdown of erythrocytes under conditions of heat stress may be the cause of the summertime decline in TEC. Our result agrees with Salem (1980) ^[15].

Total Leucocytes Count (TLC)

Tables 10, 11, and 12 show the buffaloes' total leucocyte count (thousand/µl) data from the spring and summer. In the districts of Ayodhya, Bhadowhi, and Mau, the mean ± standard error of the total leucocyte count in buffaloes during the spring was 8.42±0.03, 8.43±0.04, and 8.44±0.03 thousand/µl, respectively. In Ayodhya, Bhadowhi, and Mau districts, the mean total leucocyte count throughout the summer was 9.07±0.03, 9.07±0.04, and 9.12±0.03 thousand/µl, respectively. In all three districts, the summer season had a considerably (p < 0.01) higher total leucocyte count than the spring season. Summertime stress conditions under the effect of cortisol may be the cause of the increase in TLC, as the adrenal medulla secretes catecholamines. The rise in Total Leucocyte Count can be attributed to the mobilization of WBC from the marginal pool into the blood circulation by the catecholamines (William, 2005)^[20].

Table 1: Mean ± SE of Plasma Glucose level (mg/dl) and Blood
Urea Nitrogen (mg/dl) during Spring and Summer season in
Buffaloes in Ayodhya district of Uttar Pradesh

Week	Plasma Glucose level (mg/dl)			a Nitrogen g/dl)
	Spring	Summer	Spring	Summer
First	74.62±0.35	72.09±0.25	17.02 ± 0.19	14.71 ± 0.17
Second	74.39±0.35	72.09 ± 0.30	16.95 ± 0.19	14.56 ± 0.16
Third	74.22±0.44	72.14±0.31	16.91 ± 0.25	14.85 ± 0.19
Fourth				14.61 ± 0.19
Mean \pm SEM	74.41 ^a ±0.36	$72.10^{b} \pm 0.26$	16.93 ^a ±0.22	$14.68^{b} \pm 0.18$

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Table 2: Mean ± SE of Plasma Glucose level (mg/dl) and BloodUrea Nitrogen (mg/dl) during Spring and Summer season in
Buffaloes in Bhadohi district of Uttar Pradesh

Week	Plasma Glucose level (mg/dl)			a Nitrogen g/dl)
	Spring	Summer	Spring	Summer
First	74.40±0.30	72.20±0.24	16.55 ± 0.27	14.64 ± 0.14
Second	74.14±0.45	72.00±0.30	16.58±0.25	14.85 ± 0.19
Third	74.58±0.39	72.10±0.19	16.47±0.25	14.80 ± 0.18
Fourth	74.77±0.38	72.14±0.31	16.31±0.25	14.62 ± 0.20
Mean \pm SEM	74.47 ^a ±0.38	72.11 ^b ±0.26	16.47 ^a ±0.25	14.72 ^b ±0.18
Notes: Mean	s bearing diffe	erent supersc	ripts differ	significantly

Notes: Means bearing different superscripts differ significantly (p<0.01)

 Table 3: Mean ± SE of Plasma Glucose level (mg/dl) and Blood

 Urea Nitrogen (mg/dl) during Spring and Summer season in

 Buffaloes in Mau district of Uttar Pradesh

Week	Plasma Glucose level (mg/dl)		Blood Urea (mg	0
	Spring	Summer	Spring	Summer
First	74.05 ± 0.28	71.95±0.22	16.72±0.16	14.65 ± 0.18
Second	74.19±0.43	71.96±0.30	16.68±0.23	14.56 ± 0.17
Third	74.60±0.34	72.04±0.21	16.50±0.16	14.73 ± 0.14
Fourth	74.81±0.42	72.04±0.31	16.38±0.16	14.54 ± 0.16
Mean ± SEM	74.41ª±0.37	72.00 ^b ±0.26	$16.57^{a} \pm 0.18$	14.62 ^b ±0.16
Notes Means	bearing diff	arant supars	cripte differ	significantly

Notes: Means bearing different superscripts differ significantly (p<0.01)

 Table 4: Mean ± SE of Plasma Total Protein (g/dl) during Spring and Summer season in Buffaloes in Ayodhya district of Uttar Pradesh

Week	Plasma Total Protein (g/dl)			
vv eek	Spring Summer			
First	7.47±0.06	6.52±0.08		
Second	7.70±0.10	6.63±0.08		
Third	7.53±0.13	6.49±0.14		
Fourth	7.58±0.06	6.68±0.09		
Mean \pm SEM	7.57 ^a ±0.09	6.58 ^b ±0.10		

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Table 5: Mean ± SE of Plasma Total Protein (g/dl) during Spring
and Summer season in Buffaloes in Bhadohi district of Uttar
Pradesh

Week	Plasma Total Protein (g/dl)		
WEEK	Spring	Summer	
First	7.63±0.08	6.53±0.03	
Second	7.54±0.13	6.74±0.11	
Third	7.59±0.06	6.53±0.11	
Fourth	7.43±0.12	6.63±0.08	
Mean \pm SEM	7.55 ^a ±0.10	$6.60^{b} \pm 0.08$	

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Table 6: Mean \pm SE of Plasma Total Protein (g/dl) and Plasma
Cortisol level (ng/ml) during Spring and Summer season in
Buffaloes in Mau district of Uttar Pradesh

Week	Plasma Total Protein (g/dl)		
WEEK	Spring	Summer	
First	7.67±0.08	6.57±0.11	
Second	7.62±010	6.61±0.10	
Third	7.74±0.08	6.66±0.07	
Fourth	7.57±0.05	6.65±0.15	
Mean \pm SEM	7.65 ^a ±0.08	6.62 ^b ±0.11	

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Table 7: Mean \pm SE of Haemoglobin concentration (g/dl) and
Packed Cell volume (%) during Spring and Summer season in
Buffaloes in Ayodhya district of Uttar Pradesh

Week	Haemoglobin concentration (g/dl)			ell volume (6)
	Spring	Summer	Spring	Summer
First	11.72 ± 0.02	10.75 ± 0.04	34.73 ± 0.08	33.67±0.04
Second	11.55 ± 0.04	10.64 ± 0.06	34.63 ± 0.08	33.47±0.03
Third	11.34 ± 0.04	10.39 ± 0.08	34.29±0.03	33.16±0.02
Fourth	11.21±0.03	9.71±0.08	33.87±0.03	32.82 ± 0.03
Mean ± SEM	11.45 ^a ±0.03	10.37 ^b ±0.06	34.38 ^a ±0.06	33.28 ^b ±0.03

Notes: Means bearing different superscripts differ significantly (p<0.01)

Table 8: Mean \pm SE of Haemoglobin concentration (g/dl) and
Packed Cell volume (%) during Spring and Summer season in
Buffaloes in Bhadohi district of Uttar Pradesh

Week	Haemoglobin concentration (g/dl)		Packed Cell volume (%)	
	Spring	Summer	Spring	Summer
First	11.69 ± 0.03	10.79±0.03	34.87 ± 0.03	33.60 ± 0.04
Second	11.53 ± 0.04	10.61±0.06	34.74 ± 0.05	33.35 ± 0.05
Third	11.30 ± 0.05	10.31±0.07	34.57 ± 0.05	33.04 ± 0.04
Fourth	11.21 ± 0.03	9.69 ± 0.07		$32.53{\pm}0.06$
Mean \pm SEM	$11.43^{a} \pm 0.04$	10.35 ^b ±0.06	34.49 ^a ±0.05	33.13 ^b ±0.05

Notes: Means bearing different superscripts differ significantly (p < 0.01)

 Table 9: Mean ± SE of Haemoglobin concentration (g/dl) and

 Packed Cell volume (%) during Spring and Summer season in

 Buffaloes in Mau district of Uttar Pradesh

Week	Haemoglobin concentration (g/dl)		Packed Cell volume (%)	
	Spring	Summer	Spring	Summer
First	11.70 ± 0.03	10.77 ± 0.03	34.83±0.03	33.18±0.10
Second	11.59 ± 0.03	10.57 ± 0.06	34.70±0.04	32.98±0.12
Third	11.39 ± 0.04	10.19 ± 0.04	34.37±0.07	32.66±0.10
Fourth	11.18 ± 0.03	9.67 ± 0.08	33.31±0.12	32.29±0.10
Mean \pm SEM	11.47 ^a ±0.03	$10.30^{b} \pm 0.05$	$34.30^{a}\pm0.06$	32.78 ^b ±0.11

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Table 10: Mean \pm SE of Total Erythrocyte Count (million/µl) andTotal Leucocyte Count (thousand/µl) during Spring and Summerseason in Buffaloes in Ayodhya district of Uttar Pradesh

Week		ythrocyte nillion/µl)	Total Leucocyte Count (thousand/µl)	
	Spring	Summer	Spring	Summer
First	6.85±0.03	6.24 ± 0.04	8.26 ± 0.03	8.79±0.03
Second	6.79 ± 0.02	5.86 ± 0.03	8.34 ± 0.03	8.90±0.02
Third	6.66 ± 0.02	5.68 ± 0.02	8.45 ± 0.04	8.16±0.03
Fourth	6.54 ± 0.02	5.26 ± 0.03	8.61±0.04	9.44±0.04
Mean ± SEM	6.71 ^a ±0.02	5.76 ^b ±0.03	$8.42^{a}\pm0.03$	9.07 ^b ±0.03

Notes: Means bearing different superscripts differ significantly (p<0.01)

 Table 11: Mean ± SE of Total Erythrocyte Count (million/µl) and

 Total Leucocyte Count (thousand/µl) during Spring and Summer

 season in Buffaloes in Bhadohi district of Uttar Pradesh

Week	Total Erythrocyte Count (million/µl)		Total Leucocyte Count (thousand/µl)		
	Spring	Summer	Spring	Summer	
First	6.86±0.03	6.25±0.04	8.26±0.03	8.81±0.03	
Second	6.78±0.02	5.86±0.03	8.35±0.04	8.91±0.02	
Third	6.66±0.03	5.68 ± 0.02	8.47 ± 0.04	9.91±0.04	
Fourth	6.54±0.02	5.25 ± 0.02	8.46 ± 0.04	9.36±0.06	
Mean ± SEM	6.71ª±0.02	5.76 ^b ±0.03	8.43 ^a ±0.04	9.07 ^b ±0.04	

Notes: Means bearing different superscripts differ significantly (p < 0.01)

 Table 12: Mean ± SE of Total Erythrocyte Count (million/µl) and

 Total Leucocyte Count (thousand/µl) during Spring and Summer

 season in Buffaloes in Mau district of Uttar Pradesh

Total Erythrocyte Count (million/µl)		Total Leucocyte Count (thousand/µl)	
Spring	Summer	Spring	Summer
6.87±0.02	6.20±0.03	8.25 ± 0.03	8.83±0.04
6.78±0.02	5.83±0.03	8.35 ± 0.03	8.94±0.01
6.63±0.03	5.65 ± 0.02	8.50 ± 0.03	9.21±0.03
6.51±0.02	5.22 ± 0.02	8.67 ± 0.04	9.49±0.05
$6.70^{a}\pm0.02$	5.73 ^b ±0.03	$8.44^{a}\pm0.03$	9.12 ^b ±0.03
	Count (m Spring 6.87±0.02 6.78±0.02 6.63±0.03 6.51±0.02 6.70 ^a ±0.02	Count (million/µl) Spring Summer 6.87±0.02 6.20±0.03 6.78±0.02 5.83±0.03 6.63±0.03 5.65±0.02 6.51±0.02 5.22±0.02 6.70 ^a ±0.02 5.73 ^b ±0.03	Count (million/µl) (thou: Spring Summer Spring 6.87±0.02 6.20±0.03 8.25±0.03 6.78±0.02 5.83±0.03 8.35±0.03 6.63±0.03 5.65±0.02 8.50±0.03 6.51±0.02 5.22±0.02 8.67±0.04

Notes: Means bearing different superscripts differ significantly (p < 0.01)

Conclusion

Compared to the summer, the springtime demonstrated superior temperature adaptation in buffaloes. Summertime levels of the stress hormone cortisol were higher than those in the spring. According to biochemical assessment, the summer months have lower levels of blood urea nitrogen, plasma glucose, and total protein than the spring months. During Summertime there is decrease in hemoglobin, packed cell volume, and total erythrocyte count were as an increase in total leucocyte count was present. The current study concludes that buffalo's physio-biochemical reactions and adaptability are negatively impacted by heat stress.

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