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Research Article
Effects of Heat Stress on Haemato-Biochemical Parameters in Dairy Cows of the Chattogram Area, Bangladesh

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Abstract

Background and Objective: The livestock sector is an important component of agriculture in Bangladesh, with the dairy sector playing a key role in meeting the increasing demand for milk among the growing population. Bangladesh lies in a subtropical region characterized by a hot and humid climate, where seasonal variations in temperature and humidity may influence the health and productivity of dairy cattle. Therefore, this study aimed to evaluate the effect of heat stress on haemato-biochemical parameters in dairy cows of the Chattogram area. **Materials and Methods:** A three-month study was conducted from March to May 2022 at the Military Dairy Farm located in Chattogram Cantonment, Bangladesh. A total of 90 cows were initially selected based on their lactation status and blood samples were collected from 72 cows at the end of the study period. Routine hematological parameters and selected serum biochemical parameters were analyzed using standard laboratory methods to assess the physiological response of dairy cows to heat stress. Data were entered into MS Excel 2011, coded and analyzed using STATA/IC 14.2; descriptive statistics and one-way ANOVA were applied, with $p<0.05$ considered significant. **Results:** Hematological analysis revealed significant variation in White Blood Cell (WBC) counts among different lactation groups, with the highest WBC count observed during the 5th lactation ($11.50 \pm 3.03 \times 10^3/\mu L$). Most hematological parameters remained within the normal reference range; however, Mean Corpuscular Hemoglobin (MCH) and Mean Corpuscular Hemoglobin Concentration (MCHC) were higher than the normal range in 49% and 57% of the samples, respectively. Biochemical analysis showed that glucose, sodium and calcium levels were higher in cows during the 3rd lactation, whereas albumin and chloride were higher in the 4th lactation. Total protein, phosphorus, ALT and AST levels were highest in cows of the 5th lactation, with AST showing significant elevation. Phosphorus levels were above the normal reference range in approximately 60% of the samples. **Conclusion:** The findings indicate that ambient temperature influences the physiological condition of dairy cows, reflected by changes in hematological parameters, particularly WBC and biochemical indicators such as AST. These results suggest that heat stress may affect the health and productivity of dairy cattle, highlighting the importance of maintaining optimal environmental conditions for improved dairy production and physiological stability.

Key words: Heat stress, dairy cow, hemato-biochemical analysis, milk parameters

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The economy of Bangladesh mostly depends on her agriculture. In 2020-2021, the contribution of agriculture was 12.09%[1]. Livestock plays an important role in nutrition directly through the consumption of animal products and by-products. In 2020-2021, livestock's contribution to Gross Domestic product (GDP) was 1.86%[1]. The dairy sector is one of the most important parts of the livestock sector in Bangladesh. To meet the demand for milk and meat for the large population in Bangladesh, the number of dairy farms rearing high-yielding dairy cows is rising gradually. Almost 25% of the local demand is met by cattle from across the border[2].

The optimum environmental temperature for lactation depends on species, breed and degree of tolerance to heat or cold[3]. Hemato-biochemical parameters have been used to identify the effect of heat stress. Blood parameter profiles as animal response indicators can serve as the basis for the diagnosis, treatment and prognosis of diseases. Variations in haemato-biochemical parameters are found to be related to changes in ambient temperature and the temperature-humidity index, although within the physiological range for cattle. Therefore, the seasonal variations may influence the haemato-biochemical profile of cattle[3]. Hence, the research work on the effect of heat stress on the blood profile of dairy cattle on dairy farms was conducted with the overall objective of understanding the effect of heat stress on the haemato-biochemical parameters of dairy cattle on farms in Chattogram.

MATERIALS AND METHODS

Study area and selection of cows: The present investigation was conducted at Military Farm, Chattogram, located in Chattogram Cantonment, Chattogram District. This farm was selected based on well-organized management practices and regular data keeping records. For intensive monitoring, it was easy and the farm authority agreed to provide data and samples as the research required. The farming system of the dairy unit is an intensive type and the face-in, face out both systems are adopted. The study was conducted for a period of three months, starting from March 2022 to May 2022. The rectal temperature of cows increased with elevated ambient temperature, especially in April and May. Cows in their 3rd (101.90±0.3°F) and 5th (101.99±0.4°F) lactation exhibited increased temperatures in April, whereas cows in their 4th lactation reached their peak in May (101.91±0.4°F). Average ambient (100.21±0.4°F) and rectal (101.82±0.4°F) temperatures peaked in May.

There were a total of 1750 cattle in the farm; among them 1500 are female and 200 are male. Among the 1500 females, 780 are milking cows, 320 are dry and pregnant cows and 400 are heifers. A total of 90 cross breed holstein friesian (HF) dairy cows were selected for the study with approximately similar age in each parity and each parity was consisting of thirty cows. They were in 3rd, 4th and 5th parity and Body Condition Score (BCS) was the same. The cows were ensured with unique identification number by proper tagging. Parity wise cow's blood profiles were recorded for three months, from March 2022 to May 2022.

Haemato-biochemical analysis and data collection: The blood sample was collected in the month of May for hematological analysis. The blood samples were taken from all experimental cows in the experimental areas. The blood samples were shifted to the Department of Physiology, Biochemistry and Pharmacology, Chattogram Veterinary and Animal Sciences University (CVASU). Approximately 5 mL of blood was aseptically collected in the hottest time of the month of May, from 1200 a.m. to 1400 pm, from the jugular vein of cows by proper restraining techniques. Collected 2 mL blood samples were kept in vacutainers with EDTA anticoagulant and 3 mL in vacutainers without anticoagulant for smooth coagulation and serum separation. Collected blood samples kept in a cool box for transportation from the study area to the research laboratory of the Department of Physiology. After being centrifuged in the laboratory, coagulated serum samples were centrifuged using the Vacutainer centrifuge machine at 3000 rpm for 15 minutes. The serum was separated and shifted to a Vacutainer tube micropipette.

Hematological test of the blood sample: Hematological parameters were measured using an auto hematology analyzer (Cell Pack Alpha, Japan). All samples were analyzed for Red Blood Cells (RBC), Hematocrit (Hct), Hemoglobin Concentration (Hb), Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin Concentration (MCHC), White Blood Cells (WBC) and Platelets (Plt). The obtained serum samples were stored at -20°C until analysis for biochemical test[4-6].

Biochemical assay of serum sample: Proper aseptic measures were taken at the time of serum analysis in the laboratory. Serum and all reagents were thawed by keeping in room temperature for approximately 30 minutes before the analysis. The serum samples were vortexed for mixing component of the serum uniformly. The serum glucose, Total Protein (TP), Albumin (Alb), Sodium (Na), Calcium (Ca), Phosphorus (P), Chloride (Cl) were assayed using an automated Biochemical

analyzer (Humalyzer-3000, Germany) according to directions provided by the manufacturer of the kit. Randox kits were used to determine glucose, TP, Alb, Na; Ca; Cl; Chroma test kit for P and Biorex kit for AST and ALT. There were 90 cows for study. Within three months of study, 18 cows changed their parity. Therefore, a total of 72 serum samples were collected, analyzed as described in the blood sampling protocol[4-6].

Statistical analysis: All recorded data were entered into MS-Excel 2011 or sorted out, coded then exported into STATA/IC 14.2 (Stata Corporation, USA). Descriptive statistics, including percentage, mean and standard deviation, were performed. One-way ANOVA was used to compare the continuous variables among groups of cows, months and lactation. A probability level of $p<0.05$ was considered to be significant.

Ethical approval: Ethics committee (EC) of CVASU certifies that the project carried by Sheike Rezaul Karim has met the necessary requirements of CVASU EC to carry out the project activities. The CVASU EC approval number for the project is Memo no.- CVASU/Dir(R&E)EC/2024/688/1/20 Date: 24/03/2024.

RESULTS AND DISCUSSION

The study was conducted to observe the effect of heat stress on the blood parameters of dairy cows. The blood samples were taken from all experimental cows in the experimental areas. The blood samples were collected in the month of May for hematological and biochemical analysis. The findings are being discussed in the subsequent paragraph.

Effect of heat stress on Hematological parameters of lactating cows: Hematological parameters were observed in heat-stressed cows during May. Mean±SD values of hematological parameters are given in Table 1. The values of RBC, Hb, MCV, MCH, MCHC and PLT of stressed cows among different lactation were insignificant in the month of May. The highest value of WBC (11.50±3.03×10^3/ µL), RBC (8.04±1.76 ×10^4 / µL) and Hb (10.31±1.04 g/dL) were observed in cows at 5th lactation. The study also revealed the highest value of HCT (33.41±5.172%), MCV (33.41±4.515 fL) MCH (13.93±3.195 Pg) MCHC (30.35±.623 g/dL) and PLT (343.57±132.351×10^3/ µL) in cows at 4th lactation. However, insignificant variation existed among the Mean±SD values of all hematological parameters of lactating cows, except WBC, where the variation was significant among the lactating cows. Hematological parameters of the 72 samples were observed in heat-stressed cows during May. The values of hematological parameters are given in Table 2. The values of WBC, RBC, Hb, HCT% and MCV were within the normal value range, whereas MCH, MCHC and PLT were higher in value.

Effect of heat stress on biochemical parameters: Different biochemical parameters were studied in May and variations in parameters were found in stressed cows at different lactation stages. Mean±SD values of carbohydrate and protein levels of different lactating stressed animals are given in Table 3. The highest mean±SD values of Glucose, Total Protein and Albumin were (25.05±6.39 mg/dL), (85.92±19.52 mg/dL), (40.14±5.25 g/L), respectively, in 3rd, 5th and 4th lactating stressed cows in May. Mean±SD values of Glucose, Total Protein and Albumin found in stressed lactating cows were insignificant ($p>0.05$).

The Mean±SD values of minerals in heat-stressed cows are shown in Table 3. The highest mean±SD values of Na, P, Ca and Cl of stressed animals were 135.18±10.35 mmol/L, 6.88±2.83 mmol/L, 8.97±0.99 mg/dL and 121.59±8.43 mmol/L, respectively of 3rd, 4th and 5th lactating cows in the month of May. In addition, no significant ($p>0.05$) variation among values of Na, P, Ca and Cl was observed in milking cows at different lactation periods. Table 3 also shows the Mean±SD values of enzymes in heat-stressed lactating cows monitored in May. The highest Mean±SD values of blood ALT and AST of stressed animals were 39±3.43 U/L and 89.125±18.84 U/L, respectively of 5th lactating cows in May. This study also showed a significant ($p<0.05$) variation in blood AST levels among cows at different lactation periods.

The biochemical parameters of the 72 samples were observed in heat-stressed cows during May. The values of biochemical parameters are given in Table 4. The values of Glucose, Total Protein, Albumin, Na, Ca, Cl, ALT and AST was normal value range where as p value was higher.

Heat stress affects the productive performance of dairy animals by reducing their dry matter intake (DMI), feed efficiency and milk yield[6]. There is limited study on the effects of heat stress of dairy cows' hematological and biochemical profiles exist in Bangladesh. Therefore, hematological and biochemical profiles had been used to study effect of heat stress on productivity in lactating cows. In this study, heat stress increases with an increase in ambient temperature, resulting in a concomitant increase in the body temperature of the cow[7]. However, body temperatures that exceed normal values are not ideal and cows have been shown to decrease their feed intake and heat exchange capacity accordingly which has been stated. In Bangladesh, study on the heat stress which effects of dairy cows' haematological and biochemical

Table 1: Hematological parameters of lactating cows (N = 72)

	Stages of cows (Mean±SD)			
Parameter	3rd lactation	4th lactation	5th lactation	p-value
RBC (10^4/µL)	7.72±1.05	7.54±1.16	8.04±1.76	0.551
WBC (10^3/µL)	9.33±2.20	11.46±4.31	11.50±3.03	0.040
Hb (g/dL)	10.23±1.57	9.99±1.43	10.31±1.04	0.764
HCT %	33.18±5.91	33.41±5.17	31.21±7.28	0.624
MCV (fL)	44.18±3.82	43.41±4.52	44.21±6.51	0.624
MCH (Pg)	13.13±1.07	13.93±3.20	13.18±2.04	0.395
MCHC (g/dL)	30.28±0.70	30.35±0.62	29.91±0.71	0.267
PLT 10^3/µL	317.67±111.67	343.57±132.35	337.13±188.58	0.740

Values are presented as Mean±Standard Deviation (SD). RBC: Red blood cell count, WBC: White blood cell count, Hb: Hemoglobin, HCT: Hematocrit, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, PLT: Platelet count, µL: Microliter, g/dL: Grams per deciliter, fL: Femtoliter, Pg: Picogram. A p-values were obtained using One-Way Analysis of Variance (ANOVA), with significance considered at $p<0.05$. Reference values are provided in parentheses and were adopted from references 4 and 5. N: Number of animals sampled per lactation group

Table 2: Frequency of hematological parameters of lactating cows (N= 72)

Parameter	Standard	Normal range n (%)	Higher n (%)	Lower n (%)
RBC (10^4/µL)	5-10 (8)	69 (96)	3 (4)	0 (0)
WBC (10^3/µL)	4-12 (8)	56 (78)	16 (22)	0 (0)
Hb (g/dL)	9-15 (12)	65 (90)	0 (0)	7 (10)
HCT %	25-35 (30)	58 (81)	14 (19)	0
MCV (fL)	40-48 (44)	54 (75)	9 (12.5)	9 (12.5)
MCH (Pg)	12-13 (13)	29 (40)	35 (49)	8 (11)
MCHC (g/dL)	29-30 (30)	29 (40)	41(57)	2 (3)
PLT 10^3/µL	130-330 (250)	45 (63)	25 (34)	2 (3)

N: Sample number, n: Total number of subjects µL: Micro liter, g/dL: Gram per deciliter, fL: Femtoliter, Pg: Pico gram, RBC: Red blood cell count, WBC: White blood cell count, Hb: Hemoglobin, HCT: Hematocrit, MCV: Mean corpuscular volume, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration and PLT: Platelet count

Table 3: Blood biochemical parameters in lactating cows (N=72)

	Stages of cows (Mean±SD)			
Parameter	3rd lactation	4th lactation	5th lactation	p-value
Glucose (mg/dL)	25.05± 6.39	24.89±7.52	20.725±12.64	0.263
Total protein (g/L)	82.72±11.45	84.727±13.44	85.915±19.52	0.75
Albumin (g/L)	39.8±5.01	40.14 ±5.25	39.24±7.95	0.892
Na (mmol/L)	135.18±10.35	131.04 ±8.29	129.19±9.48	0.0988
P (mmol/L)	6.43±1.03	6.46±0.90	6.88±2.83	0.6295
Ca (mg/dL)	8.97± 0.99	8.73±1.44	8.55 ±1.28	0.4002
Cl (mmol/L)	119.51±12.74	121.59±8.43	120.98± 7.99	0.7355
ALT (U/L)	33.26 ±6.50	33.08 ±2.50	39.00±3.43	0.7132
AST (U/L)	68.36 ±9.80	73.82 ±6.58	89.125 ±18.84	0.0287

Glucose (mg/dL) 45-75(60), Total protein (g/L) 66-87(75), Albumin(g/L) 38-51(45), Na (mmol/L) 125-155(140), P (mmol/L) 3.5-5.5 (4.5); Ca (mg/dL) 8-11(9.5); Cl (mmol/L) 98–120 (115); ALT (U/L) 16- 42 (30); AST (U/L) 37–120 (65) adopted from[4,5] Values are expressed as Mean±Standard Deviation (SD). Glucose: Blood glucose concentration, Total Protein: Serum total protein, Albumin: Serum albumin, Na: Sodium, P: Phosphorus: Ca: Calcium, Cl: Chloride; ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, N: Number of animals sampled per lactation group, Units: mg/dL: Milligrams per deciliter, g/L: Grams per liter, mmol/L: Millimoles per liter and U/L: Units per liter. A p-values were calculated using One-Way Analysis of Variance (ANOVA), with statistical significance considered at $p<0.05$

Table 4: Comparison of blood biochemical parameters in lactating cows (N = 72)

Parameter	Standard	Normal range n (%)	Higher n (%)	Lower n (%)
Glucose (mg/dL)	20-30 (25)	49 (68.06)	14 (19.44)	9(12.5)
Total protein (g/L)	66-87 (75)	48 (66.67)	20 927.78)	4(5.56)
Albumin (g/L)	38-51 (45)	43 (59.72)	2(2.78)	27(37.5)
Na (mmol/L)	125-155 (140)	64 (88.89)	0 (00)	8 (11.11)
P (mmol/L)	3.5-5.5 (4.5)	12 (16.67)	60 (83.33)	0 (00)
Ca (mg/dL)	8-11 (9.5)	56 (77.78)	4 (5.56)	12(16.67)
Cl (mmol/L)	98-120 (115)	44 (61.11)	28 (38.89)	0 (00)
ALT (U/L)	16- 42 (30)	61 84.72)	11(15.28)	0 (00)
AST (U/L)	37–120 (65)	70 (97.22)	2 (2.78)	0 (00)

N: Sample number, SD: Standard deviation, n: Total number of subjects, mg/dL: Milligram per deciliter, g/L: Gram per liter, mmol/L: Micro molecule per liter, U/L: unit per liter, Na: Sodium, P: Phosphorus, Ca: Calcium, Cl: Chloride, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase and N: number of animals sampled per lactation group

profiles was rare in milking cows[8]. Therefore, haematological and biochemical profiles had been used to study effect of heat stress on productivity in lactating cows[9].

Hematological parameters have been used to identify the effect of heat stress on productivity in dairy cattle. Blood parameter profiles as animal response indicators can serve as the basis for the diagnosis, treatment and prognosis of diseases[9]. Hematological parameters have been used to identify effect of heat stress on productivity in dairy cows[9]. The values of RBC, Hb, MCV, MCH, MCHC and PLT of stressed cows among different lactation were insignificant ($p>0.05$) in the month of May[10].

The highest value of WBC ($11.50\pm3.03\times10^4$/μL), RBC ($8.04\pm1.76\times10^4$/μL) and Hb (10.31 ± 1.04 g/dL) were observed in cows at 5th lactation which is similar stated with Feldman *et al.*[7]. The study also revealed the highest value of HCT (33.41 ± 5.172%), MCV (33.41 ± 4.515 fL) MCH (13.93 ± 3.195 Pg) MCHC ($30.35\pm.623$ g/dL) and PLT ($343.57\pm132.351\times10^3$/μL) in cows at 4th lactation. However, insignificant ($p>0.05$) variation existed among the values of all hematological parameters of lactating cows, except WBC, where the variation was significant ($p<0.05$) among the lactating cows. Comparison of the hematological parameters of the samples was observed in heat stressed cows during May. The values of WBC, RBC, Hb, HCT% and MCV were normal value range whereas MCH, MCHC and PLT were higher in value[10].

Biochemical parameters are found variation related to changes in ambient temperature and temperature-humidity index, although within the physiological range for cows which is found previously[10]. Different biochemical parameters were studied in May and variations in parameters were found in heat stressed cows at different lactation stages[11]. van Saun and Sniffen[5] showed that the values of carbohydrate and protein levels of different lactating stressed animals are changed due to heat stress. The highest values of Glucose, Total Protein and Albumin were (25.05 ± 6.39 mg/dL); (85.92 ± 19.52 mg/dL); (40.14 ± 5.25 g/L), respectively, in 3rd, 5th and 4th lactating stressed cows in May. The values of Glucose, Total Protein and Albumin found stressed in high-temperature-reared lactating cows were insignificant ($p>0.05$).

The values of minerals in heat-stressed cows were the study findings. The highest values of Na, P, Ca and Cl of stressed animals were 135.18 ± 10.35 mmol/L, 6.88 ± 2.83 mmol/L, 8.97 ± 0.99 mg/dL and 121.59 ± 8.43 mmol/L, respectively of 3rd, 5th, 4th and 4th lactating cows in May[12]. In addition, no significant ($p>0.05$) variation among values of Na, P, Ca and Cl was observed in milking cows at different lactation periods[12]. The study shows the values of enzymes in heat-stressed lactating cows monitored in May. The highest values of blood ALT and AST of stressed animals were 39 ± 3.43 U/L and 89.125 ± 18.84 U/L, respectively of 5th lactating cows in May[12]. This study also showed a significant ($p<0.05$) variation in blood AST levels among cows at different lactation periods. The biochemical parameters of all the samples were observed in heat-stressed of cows during May. The values of Glucose, Total Protein, Albumin, Na, Ca, Cl, ALT and ST as in normal velour range whereas p value was higher[12].

CONCLUSION

The study revealed highest ambient, cow shed and rectal temperature of cows. Haemato-biochemical parameters in dairy animals are affected by heat stress. The ambient temperature directly affects the physiology of cows, as reflected in changes in hematological parameters. The findings also indicate that ambient temperature influences the physiological condition of dairy cows, reflected by changes in hematological parameters, particularly WBC and biochemical indicators such as AST. These results suggest that heat stress may affect the health and productivity of dairy cattle, highlighting the importance of maintaining optimal environmental conditions for improved dairy production and physiological stability. Therefore, there could be a possible effect of heat stress on haemato-biochemical parameters of dairy cows and an optimum environmental temperature is necessary for the body physiology of dairy cows.

SIGNIFICANCE STATEMENT

This study is significant because it demonstrates how heat stress affects the haemato-biochemical profile of dairy cows in a subtropical, high-humidity environment. The findings provide evidence that elevated environmental temperatures can induce measurable physiological changes, particularly in WBC and AST levels, indicating altered immune and metabolic responses. These results can help veterinarians and dairy producers better understand heat stress impacts and support the development of management strategies to improve animal health, productivity and welfare under changing climatic conditions in Bangladesh and similar regions.

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