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DIURNAL CLIMATIC PRESSURE ON HAEMATOLOGY AND BLOOD BIOCHEMISTRY OF WEST AFRICAN DWARF SHEEP

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ABSTRACT

Twelve 2.5-year-old West African Dward (WAD) sheep consisting of eight (8) ewes and four (4) rams with mean body weight 19.4kg were used to study the effects of diurnal (morning and afternoon) climatic variations on the haematological and biochemical responses in WAD sheep. The animals were randomly assigned to two treatment groups with three replicates for each treatment. Blood samples ((80ml) were collected from each of the three replicates twice per day at 9am and 3pm for 12 weeks. A significantly ($p<0.05$) higher diurnal relative humidity (57.805) and wind velocity (3.03km/hr) were recorded in the morning hours, with a significantly ($p<0.05$) higher ambient air temperature (32.36^{0}C) and radiation intensity (0.87kcal) noticed during the afternoon period. Most of the climatic factors were positively ($p<0.05$) correlated with ambient air temperature, except the relative humidity 9RH) which had a negative relationship (r—o.25) with the air temperature. Most of important haematological and biochemical parameters were fairly distributed throughout the diurnal periods, except blood glucose, which was significantly ($p<0.05$) increased with increased diurnal radiation intensity and relative humidity. Ambient air temperature exhibited positive correlations with red blood cells (r=0.41), packed cell volume (r=0.50 vs 0.20) and negative correlations with white blood cells (r=-11), plasma protein (r=-0.50) and blood glucose (r=-0.61). on the contrary, RH had positive ($p<0.05$) correlations with plasma protein (r=0.97), blood glucose (r=0.50), blood coagulation time (r=0.26) and specific gravity fo plasma/whole blood (r=0.36 vs 0.280. red blood cell, haemoglogin, glucose and specific gravity of plasma/whole blood showed positive relationship ($p<0.05$) with wind velocity and radiation intensity, except packed cell volume and white blood cells which had negative correlations with radiation intensity 9r=-0.91 and 0.40). The results of this study suggested that WAD sheep have the inherent ability to tolerate vagaries of diurnal tropical climatic conditions through prompt corrective adjustments of their body haematology and blood biochemistry.

Keywords: Diurnal climate, haematological and biochemical responses West African Dward Sheep, humid tropical climate.

INTRODUCTION

In an attempt to increase animal protein available to the world teeming populace, many of climatic changes on the body physiology 9Anjum *et al.*, 1990; Umesiobi, 2000 a, b), nutrition and metabolic pathway (NRC, 1981; Allen, 1990; Baird Casu *et al.*, 1991; Umesiobi and Iloeje, 1999) and profitability (Wilson et al 1989; Mishra, 1990; Iji *et al,* 1996) of domestic animals. However, there is a paucity of information available on the impact of diurnal climatic variations on the haematological and biochemical milieu of sheep. In a few reports (Bell *et al.*, 1976; Mack and Okali, 1985; Anjum et al, 1990 and Casu et al, 19910 on the influence of microclimate on blood physiological and biochemical integrity of sheep, it was discovered that prolonged exposure to increased ambient air temperature coupled with high relative humidity resulted to constant adjustments in the blood values and biochemical profile of the animals far away from their physiological comfort zone with a concomitant retardation in the functioning of the physiological processes and their general body performance.

Although, the West African Dwarf (WAD) sheep has the inherent reputation of being hardy and well acclimatized to the humid tropics, it is very pertinent to ascertain the haematological and blood biochemical potentials of WAD sheep that may be either activated or inhibited at different climatic conditions.

The ultimate aim of this study therefore, is to determine the response of body haematology and blood biochemistry of WAD sheep to diurnal humid climate.

MATERIALS ANDMETHODS

Experimental Studies

This experiment was undertaken on 12 adult West African Dwarf (WAD) sheep consisting of 8 ewes and 4 rams aged 2.5 years with mean body weight of 19.4kg. They were kept at the sheep Research Unit of University of Nigeria Farm.

During the period of study, the various parameters were collected diurnally, in the morning (9am) and afternoon (3pm) for a period of 12 weeks, during the dry season (January _ March). The climatic variables: ambient air temperature, relative humidity, wind velocity and radiation intensity were recorded twice each day (Morning and afternoon) form maximum and minimum thermometer, a motor-aspirated psychrometer, 3-cup anemometer and gunbelanni respectively at the university of Nigeria Metereological station located within the vicinity of the experimental site.

Housing and Flock Management

The 12 animals were randomly assigned to two treatment groups with three replicates for each group. Two weeks prior to the commencement of the experienment, the animals were placed in individual pens with ad libitum access to food and water. They were fed with a supplementary concentrate of 350g/head/day. Animals were routinely treated with Piperazine adepate (pfizers, Nig. Ltd., Lagos) against internal and external

Experimental Records

Blood samples were withdrawn from each animals twice per day through the jugular vain using a 12 guage (6cm) needle, a total of 80ml of blood colleted from each of the three replicates per two treatment groups was immediately emptied into heparinized packs containing about 40mg of anti-coagulant EDTA for laboratory analyses. In each blood sample, 10ml of blood was collected into a heparinized syringe for analysis of Red Blood Cell (RBC), Packed Cell Volume (PCV), haemoglobin, White Blood Cell (WBC), Plasma protein, Bood glucose, Coagulation time, specific gravity of plasma and whole blood.

Packaged Cell Volume (PCV) is the ratio of volume of cells to the volume of plasma. The PCV was determined by the capillary haematocrit method, using the international micro-haematocrit centrifuge as described by Coghlan *et al.* (1977) who employed capillary tubes measuring 75mm x 1.0mm, and centrifuged at 10,000 r.p.m for 5min. The haematocrit value in the capillary tube was then determined by the use of a reader.

Haemoglobin concentration was determined using the Cyanmethaemoglobin method. This involve the use of a spectrophotometer to measure the light of absorption of a mixture of blood and Drabkin's solution at a wave length of 540 pm.

The hemoglobin concentration (cb g/100m) is then extrapolated as:

$$Cb = \frac{(V + 0.02)(Ab + Cs)}{\frac{0.02\ AS}{100}}$$

where Cb = Concentration of Heamoglobin in a given sample

Ab = Absorbance of the corresponding sample cuvet

Cs = Cyanmethaemoglobin concentration of the standard

V = Volume of Drabkin's solution used.

As = Absorbance of the standard cyanmethaemoglobin (Willard and John, 1970).

Erythrocyte or red blood cell (RBC) count was determined by the use of hemocytometer. In this method, 10ml of whole blood was diluted, with 2 x 10^2ml of distilled water to produce a dilution of one par of blood and 20 part of diluting fluid. The blood and fluid were thoroughly mixed in a pipette and the counting chamber containing a slide was then filled with the mixture to a depth of 0.1mm. The RBC was counted in the counting chamber made of 400 squares representing one square millimeter. The leucocyte or white blood cell (WBC) count was ascertained using the hemocytometer and a staining leucocyte diluting fluid by the procedures of Bell et al (1976).

Plasma protein was determined using the Biuret methods as described by Fu et al. (1993). This device involved the use of a spectro- photometer in the reading of absorbance of the solutions at 540 pm after incubating the mixed solution for 30minutes. The plasma protein content was then estimated using the formula:

Plasma protein gm/100ml of blood = Absorbace of unknown x gm protein standard

Absorbance of Standard 1

Blood glucose level was estimated using the Folin and Wu method (Allen, 1990; Ullman *et al.*, 1992; Potocnik and Wintour, 1996). In this method, a protein – free filtrate was first prepared by transferring exactly 1ml of blood to a dry 20ml capacity test tube mixed with 7ml of water, 1ml of $^2/_3$ NH_2 SO_4. The filtrate was later transferred to Folin and Wu sugar with 25m capacity graduated tubes and 2ml of standard sugar solutions containing 0.2 and 0.4mg of glucose was added. The concentration of glucose was thereafter determined in mg/100ml blood as shown below:

Glucose mg/100ml blood= Density of unknown x Mgglucose in standard x 100

Density of Standard 2

Blood clotting time is the time lag from the period blood was withdrawal from the animal into a syringe to the moment strands of fibrin started to form. The glass-slid method (Ullman *et al.*, 1992) was used to determine the whole blood clotting time. Specific gravity of whole blood had plasma were measured using specific gravities as outlined by Potocnik and Wintour (1996). A stock solution of copper sulphate of specific gravity 1.00 was prepared by making up a 17% $Cuso_4$ solution. Different specific from the test stock using distilled water. The specific gravities of the blood or plamas were then determined by adding 1 or 2 drops of the sample into each of the prepared solutions of unknown specific gravities. Any of the solutions in which the blood or plasma sample floated was recorded to have equal specific gravity with the sample.

Statistical Analysis

The relationships between the diurnal climatic variables and the animal haematologicla and biochemical characteristics were determined using correlation analysis (Little and Hills, 1987). Analysis of variance was used to test for significant effects between the various parameters in accordance with the procedures outlined in Statistical Analysis System (SAS) (1990) programme, and treatment means were differentiated using Duncan New Multiple Range Test procedure (Snedecor and Cochran, 1980).

Results and discussion

The diurnal climatic conditions of the experimental site are shown in Table 1. The effects of diurnal period on the body haematology and biochemical responses of West African Dwarf (WAD) sheep are presented in Table 2. A correlation of certain climatic factors with the haematological and biochemical responses of WAD sheep is summarized in Table 3.

Diurnal Climatology

A lower ambient air temperature (25.52^{0}C) was recorded at 9 am (morning hours) with a statistically ($p<0.05$) higher mean value (32.36^{0}C) observed at 3pm (afternoon hours). However, a higher relative humidity RH of 57.8% was noticed in the morning period with a lower RH (49.4%) obtained during the afternoon hours. There existed a negative relationship between ambient air temperature and the RH ($r = 0.25$).

An average wind velocity of 3.03km/hr was recorded during the morning period with a higher value (5.57km/hr) noted at 3pm. There were a positive correlations between wind velocity and the ambient air temperature ($r=0.60$), and a negative correlation with the relative humidity ($r=0.13$). The mean radiation intensity was higher in the afternoon (0.95kcal) than the morning (0.87kcal) values. However, radiation intensity was positively ($p<0.05$) related with the ambiet temperature ($r=0.04$), and wind velocity ($r=0.53$) and a negative relationship with the relative humidity ($r=-0.29$). These records are in consonance with the reports of McDowell (1989) and Mishra (1990) who noticed that as the environmental temperature increased, other climatic variables assumed significance in maintaining the animal's homeostasis. Hence, the interactions (rather than solation) effects of these environmental factors act in concert on the haematological and biochemical characteristics of the animal (Duncan, 1993).

TABLE 1: Diurnal climatic conditions of the experimental site.

	DIURNAL PERIOD			
	MORNING		AFTERNOON	
CLIMATIC VARIABLES	**Mean**	**SE**	**Mean**	**SE**
Ambient air temperature ^{0}(C)	25.52^a	0.98	32.36^b	0.58
Relative humidity (%)	57.80^a	14.36	49.40^b	15.76
Wind velocity (km/hr)	5.57^a	1.34	3.03^b	0.71
Radiation intensity (kcal)	0.87^a	0.04	0.95^b	0.04

Within each row, figures differently superscripted are significantly ($p<0.05$) different.

Body Haematology

Red blood cells (Erythrocytes: The mean read blood cell (RBC) values of 4.73 x 10^6/mm^3 was obtained I the morning and 4.73 x 10^6/mm^3 recorded during the afternoon hours. Although, no significant ($p>0.05$) differences were observed between the morning and afternoon RBC records, there were positive correlations between RBC and ambient air temperature ($r=0.14$), wind velocity ($r = 0.14$), except relative humidity, which had a negative relationship with the RBC ($r = 0.14$). This result is in accord with the observations of Horton (1978) and Ullman et al (1992) who recorded increases in RBC due to increased radiation and stress associated with constant haemorrhage in the animals.

Packed call volume (Haematocrit). The evening period has a non-significant ($p>0.05$) higher mean packed cell volume (PCV) of 25.65% than that of the morning (24.89%) value. The PCV was significantly ($p<0.05$) affected by the ambient temperature and wind velocity. PCV was also positively correlated with ambient air temperature ($r = 0.03$) and wind velocity ($r = 0.02$). On the contrary, PCV had negative correlations with relative humidity ($r = 0.14$) and radiation intensity ($r = -0.19$). The significant ($p< 0.05$) increase recorded in PCV might have been aggravated by heat stress caused by constant increase in ambient temperature, which according to Anjum, et al (1990) and Aganga (1992) predisposes the animals to blood related diseases. Haemoglobin mean value of haemoglobin (Hb) was 10.09gm% in the morning and non-significantly ($p>0.05$) higher during the afternoon period (10.81mg%). The Hb level of the animals has positive correlations with ambient air temperature ($r=0.17$), wind velocity ($r=0.02$) and radiation intensity ($r=0.23$), but negatively related with relative humidity ($r=17$). The significant increase noticed in Hb level was caused by the persistent increases in the ambient temperature whose effects were complemented by the corresponding

increase in wind velocity and radiation intensity. This observation is agreement with the findings of Oduye (1976) and Ullman *et al* (1992) who reported a corresponding increase of Hb with ambient air temperature, mostly when the animals are subjected to field trials. White blood cells (Leucocytes). As shown in Table 2, no statistical ($p>0.05$) differences were noticed in white blood cell (WBC) count between the morning (8.74 x $10^3/mm^3$) and afternoon (8.82 x $10^3/mm^3$) values. Negative relationships were also observed between the WBC and ambient air temperature (r=-0.11), relative humidity (r=-0.02), wind velocity (r=-0.04). This record is in line with the reports of McDowell (1972), Singh and Singh (1992) and Iji *et al* (1996) who established that an erratic internal climatic condition of sheep induced an adaptive functioning of the basic physiological mechanisms and blood biochemistry, mostly te mobility of WBC which constitute the firs line of defense against infection or stress.

Table2: Effects of diurnal period on the haematology and biochemistry of West African Dwarf sheep

	DIURNALPERIOD			
	MORNING		AFTERNOON	
Parameters	Mean	SE	Mean	SE
Red blood cells ($x10^6$)/mm^3	4.30	0.26	4.73	0.37
Packed cell volume (%)	24.89	0.83	25.56	0.80
Haemoglobin (gm%)	10.09	0.35	10.81	0.58
White bood cells ($x10^3$)/mm^3	8.74	0.87	8.82	0.58
Plasma protein (mg%)	8.57	0.22	8.61	0.18
Blood glucose (mg%)	69.44[a]	13.40	58.81[b]	12.16
Coagulation time (min)	3.22	0.35	2.87	0.27
Specific gravity of plasma	1.041	0.0008	1.042	0.0013
Specific gravity of whole blood	1.059	0.0009	1.060	0.0009

a, b: With each row of parameter, figures differently superscripted are significantly ($p<0.05$) different.

Plasma protein. Diurnal climatic change did not cause any significant ($p>0.05$) alternation on the plasma protein. However relative humidity and radiation intensity had positive correlations with plasma protein (r=0.97 and 0.37) respectively, while negative relationships existed between plasma protein and ambient temperature (r=0.05) and wind velocity (r=-0.41). The increased plasma protein observed with decreased ambient air temperature and wind velocity may be due to the complementary effects of increased radiation intensity and relative humidity (see Table 3). The non-significant ($p>0.05$) change in plasma protein in both morning and afternoon hours is in accord with the views of Block et al (1987), Payne (1990) and Duncan (1993) who reported that plasma protein was noticed to be remarkably constant diurnally in climatically normal adult sheep.

Blood glucose. Mean blood glucose level of 69.44mg% was recorded in the morning hours with a significantly ($p<0.05$) lower value (58.81mg%) observed in the afternoon period. Blood glucose had positive relationships with relative (r=0.05) wind velocity (r=0.03) and radiation (r=-0.61). This observation is in conformity with the reports of NRC (1981) and Adams and Sanders (1992) who in their various studies noted significantly ($p<.05$) combined effects of increased radiation intensity and relative humidity on sheep blood glucose level.

Special gravity of plasma and whole blood showed significantly ($p<0.05$) relationship with all the climatic variables.

CONCLUSION

The results of this study indicated that most of the haemeters were not significantly ($p>0.05$) affected by any of the diurnal climatic periods, except the blood glucose which was significantly ($p<0.05$) increased over the diurnal hours. However, the animals struggled to maintain their

body steady-state by making non-significant changes in their body haematology and biochemistry to the various climatic variations. This therefore, suggests that WAD sheep has a characteristic ability to tolerate extremes of climatic conditions. This is made possible by the animals' baemotological and biochemical corrective mechanisms they elicited to recuperate from stressful condition of their environment.

Table 3: Correlations between the climatic factors and the body hematology and biochemistry of West African Dwarf sheep.

PARAMETERS	Ambient air temp (°C)	Relative humidity (%)	Radiation intensity (Km/hr)	Wind velocity (Kcal)
Ambient air Temperature (°C)	-	-	-	
Relative humidity (%)	-0.25	-	-	-
Wind velocity (km/hr)	0.06*	-0.13	-	-
Radiation intensity (Kcal)	0.04	-0.29	0.53*	-
Red blood cells ($x10^6$)/mm^3	0.14	-0.42	0.04	0.14
Packed cell volume (%)	0.03	-0.14	0.02	-0.19
Heamoglobin (gm%)	0.17	-0.17	0.02	0.23
White blood cells I($x10^3$)/mm^3	-0.11	-0.02	-0.13	-0.04
Plasma protein (gm%)	-0.05	0.97*	-0.41	0.37
Blood glucose (mg%)	-0.61	0.05	0.03	0.80**
Coagulation time (min)	-0.04	0.26	-0.16	0.02
Specific gravity of plasma	0.02	0.28	0.1	0.42
Specific gravity of whole blood	0.05*	0.36	0.13	0.41

*$(p<0.05)$
**$(p<0.01)$

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