

THE EFFECT OF SEASONAL VARIATIONS ON BODY AND VASCULAR FLUIDS OF BUFFALOES UNDER EGYPTIAN CLIMATIC CONDITIONS.

Mona A. El-Khashab; Ali R. Abd El-Rhman and Amr, M. Haider
Faculty of Agriculture, Department of Animal Production, Fayoum University.

ABSTRACT

The present study was carried out using twelve non lactating non pregnant buffaloes at the experimental research station, College of Agriculture, Fayoum University. All animals were 4-5 years old, and were kept under shad with a galvanized steel shed. Seasonal effects on thermoregulatory parameters, blood biochemical and body vascular fluids accompanied with hormonal study were aimed to the present study. The experimental periods starting from June 2004 up to May 2005 which include the four season of year (summer, autumn, winter and spring).

Rectal temperatures (RT), respiration rate (RR) and pulse rate (PR) were measured. Two blood samples were taken one of them to determine some hematological parameters and other to obtain serum. Haematocrite ratio (Ht %) and blood hemoglobin content (Hb, g %) were determined. serum total protein, albumin, globulin, aspartic and alanine transaminase activity (AST and ALT), serum sodium (Na), potassium(K), chloride (Cl), aldosterone and cortisol hormones were measured. Total body fluid (TBF, L) extracellular fluid (ECF) and interstitial fluid volume (ISF), moreover plasma (PV) and blood volume (BV) were determined.

There was a significant seasonal difference ($P < 0.01$) in RR, PR. Total protein, albumin and globulin were increased significantly ($P < 0.01$) in winter than in summer season. While liver enzymes, concentrations of Na, K and Cl were decreased significantly in winter than summer season.

The highest value of aldosterone level was in winter and the lowest value was shown in summer season. The absolute and relative values of TBF, ECF, ISF, PV and BV were greater significantly ($P < 0.01$) in summer than other seasons.

Key words: Buffaloes, Blood constituents, Season of year, Body Fluid, Hormones.

INTRODUCTION

The seasonal variations range of ambient temperature is very wide so that the physiological mechanisms of the animal are always endeavoring to cope with seasonal fluctuations in the environmental conditions. This tedious state is reflected on the productive and reproductive performance of the animal as well as on its vitality and viability in accordance with its capability, efficiency and ease of fulfilling the proper physiological counteraction (Yousef and Johnson, 1985).

Water buffaloes (*Bubalus bubalis*) are the main dairy animal in Egypt and other tropical countries which can tolerate the adverse environmental condition and the poor managerial and feeding conditions that prevail in such area. Moreover buffaloes are known to differ from other domestic ruminants both in heat tolerance and water requirements. Hence physiological response of buffaloes to environmental factors has been the focus of some interest during the last four to five decades (Ashour, 1990).

The physiological reaction of *Bubalus bubalis* towards fluctuations in climatic conditions occur in two synchronized directions; control of thermolysis which includes heat output from the body and the other direction is controlling the thermogenesis through heat input by metabolic activities, furthermore, feed and body fluid compartments including extracellular fluid, interstitial fluid, intracellular fluid, circulatory fluid and the major mineral pools are essential for the maintenance of homeostasis (Shebaita, 1993).

It is well known that the total body fluid is distributed between two major compartments, intracellular and extracellular fluid. The later compartment is divided into interstitial and vascular pools (El-Banna, 1970). In ruminants the volume venous rumen keeps a greater proportion of body water which has to be given particular consideration. There are continuous exchange of water between the body compartments and regions to maintain ionic and osmotic balances. On the other hand water mobilization is achieved in response to the external environmental conditions, particularly water and ions availability. The response to environmental condition is spontaneous and fast in vascular, interstitial and rumen water rather than the intracellular water, moreover, the volume of this intracellular water is maintained stable while the volume of water in the former compartments is flexible in accordance with the homeostatic balance in the body (Ashour 1990 and Hamed, et. al., 2007).

The main objective of this experiment was to study the effect of seasons of the year under Egyptian conditions on body fluids, adaptive parameters, blood hematology, serum metabolites and body pools of minerals. Aldosterone and cortisol hormones levels were also measured during the four seasons of the year in Egypt under EL Fayoum condition.

MATERIALS AND METHODS

Experimental animals:

A total number of 12 non lactating-non pregnant buffaloes were chosen from the herd of the experimental station of the department of animal science, Faculty of Agriculture at El-Fayoum University.

The averages body weight in summer, autumn, winter and spring were 363.8 ± 20.4 , 383.4 ± 21.2 , 390.2 ± 20.3 and 386.2 ± 17.2 Kg respectively. All animals were 4-5 years old and free from disease and behavioral abnormalities during the experimental period.

Management and feeding:

The buffaloes were kept under shad with a galvanized steel shed.

The total daily requirement was calculated for each animal by the knowledge of the average live body weight according to NRC, 1988.

In winter and spring, the animals were fed on Egyptian clover (*Trifolium alexandrinum*) and clover hay in summer and autumn, along with concentrate mixture and wheat straw. Water was provided ad-libitum twice daily during the experiment.

Experimental Design:

The experimental period starting from June 2004 up to May 2005 to know the effect of seasons (summer, autumn, winter and spring) on the parameters under study.

Methods of determinations:

Thermoregulatory parameters were recorded weekly for each animal. Rectal temperature (RT°C) was measured using a standard clinical thermometer inserted into the rectum approximately three inches after one minute the measurement was recorded to the nearest 0.1 C°. Respiration rate (RR/min) was counted from movement of flank in one minute using stop watch. Pulse rate (PR/min) was taken by placing the hand gently on the coccygeal artery and counting the pulse for a minute. All thermoregulatory parameters were taken at 10:00 a.m. from fasting animals for 12 hours.

Two blood samples from each animal monthly were collected in two test tubes. One of them containing heparin as anticoagulant to determine some hematological parameters and the other test tube was used to obtain blood serum which stored frozen at - 20°C until analysis.

Haematocrite ratio (Ht %) was determined according to Wintrobe method. Blood hemoglobin content (Hb, g / 100 ml) was determined according to Makaren, (1974) and total blood hemoglobin content in gm (To. Hb, g) and as fraction of body weight (To. Hb, g/Kg. BW), were also calculated.

The determination of total protein (g/dl) was performed by using test kits (STANBIO) according to Cannon, (1974). Albumin (g/dl) was measured by test kits (Diamond Diagnostics Egypt) according to **Beng and Lim (1973)**. Globulin concentrations were calculated by the subtraction of albumin from total protein. Albumin / Globulin ratio was also calculated.

Aspartic-transaminase (AST U/L) and Alanine- transaminase (ALT U/L) activity in serum were measured calorimetrically using test kits combination provided by Diamond Diagnostics, according to **Reitman and Frankel (1957)**.

Serum sodium (Na mmol/L) and potassium (K mmol/L) were measured colorimetrically using test kits combination provided by Biodiagnostic, according to **Sunderman and Sunderman (1958)** for potassium and **Trinder (1951)** for sodium. Chloride (Cl mmol/L) was measured colorimetrically using test kits combination provided by SPINRE ACT, S.A. SPAIN according to **Burtis (1999)**.

Serum aldosterone and cortisol were measured using commercial radioimmunoassay (RIA) kit of Diagnostic products corporation (DPC) Los Angeles, USA. according to **Mayes, (1970)** for aldosterone and **DSL-2100, Texas, USA** according to **Kreiger, (1975)** and **Migeon and Lanes, (1990)** for cortisol.

Total body fluid (TBF, L) as absolute and relative values to body weight (TBF, ml/ Kg and TBF, L%) were estimated using Antipyrine- space method as described by **Brodie, et. al., (1949)**. Extracellular fluid (ECF) was measured using sodium thiosulfate method according to **Cardozo and Edelman, 1952**. Total plasma volume (PV) was determined by the method of **Davis and Isenberg, (1953)** using the Evan's blue dye (T-1824). Blood volume (BV) was calculated by the equation of **Pandy and Roy, 1969**, where: $BV (ml) = PV (ml) \times 100 / (100 - Ht)$.

Interstitial fluid volume (ISF) was calculated by deducting the PV from ECF. Intracellular fluid volume (ICF) calculated by deducting the ECF from TBF.

Statistical analysis:

Statistical analysis were conducted according to **SPSS, (1997)** included analysis of variance, **Duncan's, (1955)** multiple range and simple regression.

RESULTS AND DISCUSSION

Seasonal Effects on Certain Thermoregulatory Parameters:

It was observed from Table (1) that there was a significant seasonal difference ($P < 0.01$) in the RR and PR which increased from winter to summer by about 36.01 % and 9.17 % respectively while no significant difference in RT and the difference between summer and winter was 1.26%. So the seasonal average of rectal temperature varied slightly, this proves that the great efficiency of adaptation of Egyptian buffaloes in the long term seasonal variations under the special housing conditions these results were agreed with the results of Ashour, (1990) and Marai, *et al.*, (1997). There was no significant difference between the average value of the respiration rate under summer and autumn condition while the average value of the spring was lower significantly ($P < 0.01$) than the value of the summer by 5.51.

Table (1): Effect of season on Thermoregulatory parameters and serum constituents in Buffaloes (Means \pm SE).

Parameters	Season			
	summer	Autumn	Winter	Spring
RT °C	38.28 \pm 0.031	38.01 \pm 0.022	37.81 \pm 0.027	37.94 \pm 0.023
RR / min **	30.62 \pm 0.520 ^a	30.43 \pm 0.542 ^a	22.50 \pm 0.348 ^c	29.02 \pm 0.439 ^b
PR / min **	63.57 \pm 0.371 ^a	60.11 \pm 0.614 ^c	58.23 \pm 0.341 ^d	61.76 \pm 0.577 ^b
T.protein,g/dl **	7.59 \pm 0.080 ^b	7.66 \pm 0.050 ^b	7.97 \pm 0.066 ^a	7.95 \pm 0.067 ^a
Albumin,g/dl **	3.81 \pm 0.049 ^b	3.54 \pm 0.048 ^c	3.98 \pm 0.056 ^a	3.84 \pm 0.059 ^b
Globulin,g/dl **	3.78 \pm 0.062 ^b	4.12 \pm 0.051 ^a	3.99 \pm 0.062 ^a	4.11 \pm 0.071 ^a
A/G **	1.02 \pm 0.020 ^a	0.87 \pm 0.020 ^c	1.01 \pm 0.026 ^{ab}	0.95 \pm 0.027 ^b
AST U/L **	94.94 \pm 3.316 ^a	62.99 \pm 2.017 ^c	77.94 \pm 2.323 ^b	66.53 \pm 2.266 ^c
ALT U/L **	15.74 \pm 0.611 ^a	15.70 \pm 0.395 ^a	11.77 \pm 0.381 ^b	14.82 \pm 0.547 ^a
Na mmol/L **	140.59 \pm 1.340 ^a	123.64 \pm 2.304 ^{bc}	118.81 \pm 2.284 ^c	141.03 \pm 1.471 ^a
K mmol/L **	6.23 \pm 0.247 ^a	5.82 \pm 0.169 ^{ab}	5.59 \pm 0.216 ^b	5.86 \pm 0.201 ^{ab}
Cl mmol/L **	116.58 \pm 1.971 ^a	112.96 \pm 0.262 ^a	107.81 \pm 1.793 ^b	114.45 \pm 0.582 ^a

Average in the same row having different superscripts differ significantly * $P < 0.05$, ** $P < 0.01$. RT: rectal temperature; RR: respiration rate; PR: pulse rate; A/G: albumin globulin ratio; AST and ALT: aspartic and alanine transaminase; Na: serum sodium; K: serum potassium; Cl: serum chloride.

The great rise in air temperature in summer increases the heat load on the animal thus evoking rise in body temperature which increase the activities of hypothalamus to stimulate the respiratory center which induces increase in the first reaction observed in cattle and buffaloes when exposed to environmental temperature above the thermoneutral zone is an onset rise in respiration rate (Yousf and Johnson, 1985). This response was found to ensure by direct heat stimulation of the peripheral receptors rather than by a raise in blood temperature and it sustains in spite of the continued fall in deep-blood temperature (Bligh, 1975). These receptors stimulate the hypothalamus by transmitting nervous impulses to the heat center in the hypothalamus, the cardio-respiratory center is stimulated to send impulses to the respiratory musculature to increase the respiratory activity with increasing environmental temperature and respiration rate continued to rise due to a direct stimulation of the central thermal receptors (Findlay and Ingram, 1961). At this level the respiratory muscles reach a threshold of maximum activity and thus the respiration rate cannot further increase. The significantly increase in respiration rate under high ambient temperature is that it enables the animal to dissipate heat by

THE EFFECT OF SEASONAL VARIATIONS ON BODY AND VASCULAR... 21

vaporizing the high moisture content of respired air, which accounts for about 30% of the total heat dissipation (McLean, 1963).

Table (2): Regression analysis for thermoregulatory parameters and serum constituents on ambient temperature (°C) for all seasons of year.

Parameters	a	b	r	R ²
RT °C	37.294	0.0164	0.4309	0.1857***
RR/ min	0.4516	0.0179	0.6054	0.3665***
PR/ min	52.018	0.2131	0.2618	0.0685***
T. protein, g/dl	8.206	- 0.0170	0.2226	0.0497**
Albumin, g/dl	3.967	-0.0072	0.1158	0.0134 ^{ns}
Globulin, g/dl	4.275	-0.0113	0.1616	0.0261*
A/G	0.939	0.0010	0.0369	0.0013 ^{ns}
AST U/L	64.048	0.4757	0.1417	0.0201*
ALT U/L	9.597	0.2023	0.3428	0.1175***
Na mmol/ L	99.314	1.3056	0.5063	0.2563***
K mmol/ L	4.896	0.0403	0.1756	0.0308*
Cl mmol/ L	101.524	0.4706	0.3013	0.0908***

*P < 0.05, ** P < 0.01, ***P < 0.001, ns: not significant.

Equation in the form $Y = a + bx$, where Y = any parameter study (dependent variable), a = intercept of the characteristics, b = regression coefficient for parameter on ambient temperature (°C), x = independent variable, r = coefficient of correlation, R² = coefficient of determination.

RT: rectal temperature; RR: respiration rate; PR: pulse rate; A/G: albumin globulin ratio; AST and ALT: aspartic and alanine transaminase; Na: serum sodium; K: serum potassium; Cl: serum chloride.

Concerning with the increasing value of PR/min in summer than other seasons this may be due to the increase of ambient temperature and the diurnal variations of PR (Ashour, 1990).

It was interesting to note that the significant positive relationship (P<0.001) between ambient temperature and thermoregulatory parameters as shown in Table (2).

Seasonal Effect on Serum Biochemical Constituents:

Serum analysis results showed that there was significant effect (P<0.01) of season on serum total protein g/dl, albumin, g/dl and globulin, g/dl as shown in Table 1. They were significantly low during summer months as compared to levels of winter. On the other hand, there was statistically insignificant difference between summer and autumn months and between winter and spring months concerning total protein. Results Table (1) also revealed that there was a significant (P<0.01) differences between blood serum albumin in summer, winter and autumn months while there was no significant differences between summer and spring months.

Results also indicated that albumin/globulin ratio was significantly (P<0.01) difference between summer, autumn and spring values. The decrease in serum protein and its fraction which was found in summer months may be attributed hemodilution effect (El-Marsy, *et al.*, 2001), because of the increase of water consumption, blood volume (Kamal, *et al.*, 1993) and body water content (Kamal, *et al.*, 1972) mainly increased in heat stressed animals. Moreover, the increase in cortisol level which accompanied with enhancing tissue destruction may be involved in such decrease of blood protein in heat stressed animals (El-Nouty, *et al.*, 1978).

The significant negative relationship ($P < 0.01$ and $P < 0.05$) was recorded between total protein, albumin and ambient temperature (Table 2).

Results, (Table 1) also indicated that during hot summer conditions the level of AST and ALT enzymes significantly increased ($P < 0.01$) in the serum of the buffaloes as compared to its levels during cool-winter conditions. The average values of AST and ALT U/L in summer were 94.94 and 15.74 U/L while they were 77.94 and 11.77 U/L in winter respectively. The change due to summer were +21.82 and +33.75 % in AST and ALT than in winter. Moreover there was no significant difference between autumn and spring in serum AST U/L levels and ALT U/L, where the two enzymes take the same manner in the two seasons. True enough, there was positive relation between liver enzymes under study and ambient temperature ($^{\circ}\text{C}$), this relation was highly significant (Table 2).

Seasonal Effect on Osmosis Response:

Blood serum concentration of Na^+ and K^+ and Cl^- were increased significantly ($P < 0.01$ and $P < 0.05$) with the increase of environmental temperature, they were higher in summer than in winter. There was no significant difference between the average value of the serum concentration of Na^+ mmol/L under summer and spring months, and / or between winter and autumn. The summer value was higher than the average of autumn and winter by 13.71 and 18.33 % respectively (Table 1).

The increased Na^+ concentration under hot condition in apt to increase the plasma osmolarity which results in drainage of water from the other body fluid compartments finally, the plasma volume and extracellular fluid increase greatly. On the other hand increase the continuous supply of water for vaporizing as the main mechanism for temperature balance (Ashour, 1990). Moreover the increase of Na^+ level in summer may help to maintain the moral osmolarity, also the low Na / K ratio in urine represents that the amount of Na^+ excreted in urine was very low compared to K^+ , this case will help to maintain optimum high concretion of Na^+ in plasma necessary to fulfill the proper biological function of the physiological system as well as the general osmolarity. Ashour, *et. al.*, (2001) reported in fistulated male buffaloes that rumen Na^+ and K^+ concentrations decreased under hot conditions. These decreases were accompanied with proportional increase in blood plasma.

The significant positive relationship ($P < 0.01$ and $P < 0.05$) was recorded between Na , K , Cl and ambient temperature (Table 2).

Seasonal Effects on Hematological Parameters:

Blood haematocrite (Ht %) value and hemoglobin concentration (Hb, g %) and Total Hb, g/kg body weight were significantly affected by the different climatic season (Table 3). While the Total Hb, g was not significantly affected. Thus, Ht % value was higher in winter season (36.63 ± 0.387) and autumn (36.56 ± 0.359 %) than the values of the summer (34.90 ± 0.313) and spring (34.73 ± 0.466). The decrease in haematocrit value % during summer season when animals subjected to elevated ambient temperature may be due to a hemodilution effect where more water is transported in the circulatory system for evaporative cooling, also this decrease in Ht % during summer months was a hemodilution effect of the greater increased in blood volume which associated with high environmental temperature (Beltagy, 1990). The increase in seasonal environmental temperature was accompanied by a decrease in Ht % values in summer than in winter in accordance with the opposite trend of change in plasma volume. Moreover, Ashour (1993) in Egyptian male buffaloes he found that there were slight decreases in the Ht % values due to the effect of stress solar radiation. The decrease in Ht value not only due to change in plasma volume (hemodilution) but also due to change in circulation RBCs in the

systemic circulation, most probably by increasing storage in spleen. Contraction of the spleen occurs when more RBCs are needed (Reece, 1991). It is interesting that solar radiation induced reduction in the concentration of RBCs circulation in the blood, an attempt to reduce O₂ carriage to depress the metabolic rate in this case of heat stress.

Concerning Hb, g %, summer season was responsible for decreasing of it. Hassan and El-Nouty (1985) in buffaloes and cross-bred (Egyptian × Holstein) heifers found that their blood Hb was higher in winter and spring than in summer and autumn. Similar trends have been reported by Marai, *et. al.*, (1997) in lactating Friesian and Holstein cows.

The significant negative relationship ($p < 0.01$ and $P < 0.05$) was recorded between Ht%, Hb, g % and ambient temperature (Table 4).

Table (3): Effect of Season on Haematocrite, Hemoglobin, Aldosterone and Cortisol hormones in Buffaloes (Means ± SE).

Parameters	Season			
	summer	Autumn	winter	Spring
Ht % **	34.90 ± 0.313 ^b	36.56 ± 0.359 ^a	36.63 ± 0.387 ^a	34.73 ± 0.466 ^b
Hb, g % **	10.82 ± 0.136 ^d	11.57 ± 0.144 ^c	12.66 ± 0.188 ^a	12.02 ± 0.197 ^b
To.Hb,g	2780.28 ± 129.20	2669.82 ± 155.23	2873.51 ± 183.12	2673.88 ± 176.08
To.Hb,g/Kg.BW**	7.90 ± 0.139 ^a	6.95 ± 0.081 ^{b,c}	7.17 ± 0.130 ^b	6.79 ± 0.134 ^c
Aldosterone, pg/ml**	17.92 ± 0.693 ^c	21.00 ± 0.223 ^b	26.69 ± 0.476 ^a	21.26 ± 0.567 ^b
Cortisol, ng /ml**	11.11 ± 0.099 ^a	9.72 ± 0.125 ^b	9.15 ± 0.149 ^c	9.92 ± 0.085 ^b

Average in the same row having different superscripts differ significantly * $P < 0.05$ ** $P < 0.01$. Ht%: haematocrite ratio; Hb, g%: hemoglobin content; To.Hb,g : total hemoglobin content; To.Hb,g/Kg.BW :total hemoglobin gm/ Kg live body weight.

Seasonal Effects on Blood Hormones Levels:

Blood serum aldosterone concentration pg/ml and cortisol ng/ml were affected significantly ($P < 0.01$) by the season of the year (Table 3). The highest value of aldosterone level was in winter 26.69 pg / ml then it slightly decreased in spring 21.26 pg / ml, and the lowest value was shown in summer season which being 17.92 pg/ml. It is apparent from these results that aldosterone level decreased greatly under sustained hot conditions in summer. Ashour, (1990) in buffaloes males, found that the seasonal difference in aldosterone concentration was very obvious in winter morning which being 22.6 pg / ml, while the average in summer morning was 13.1 pg / ml. Yousef and Johnson (1985) concluded that the decreased blood aldosterone levels under hot conditions irrespective of the low Na⁺ levels in cattle and reindeer remains a mystery.

Moreover, significant negative relationship ($P < 0.01$) was recorded between the ambient temperatures (°C) and aldosterone.

Cortisol concentration in summer season was 11.11 ng/ml and the lowest values of cortisol concentration was in winter 9.15 ng/ml. Moreover there was no significant difference in cortisol concentration between autumn and spring. Habeeb, *et. al.*, (2001) found that there are decreasing in cortisol level by 13.6 % in winter season than summer one in lactating Friesian cows. Cortisol as glucocorticoids hormone has a proteolytic action and its level increased in the heat stressed cattle and buffaloes. Stress increased secretion of cortisol from the adrenal cortex in cattle; one of the most important benefits of increased glucocorticoid secretion in stress concerns the maintenance of flight response with essential metabolites by diverting

glucose metabolism from muscle to the brain and other tissues. (Mudron, *et. al.*, 2005).

Significant positive relationship ($P < 0.001$) was recorded between the ambient temperatures ($^{\circ}\text{C}$) and cortisol.

Table (4): Regression analysis for hematological parameters, some hormones, body fluids, plasma and blood volume on ambient temperature ($^{\circ}\text{c}$) for all seasons of year.

Parameters	a	b	R	R ²
Ht %	37.899	- 0.0894	0.2124	0.0451**
Hb,g %	14.004	- 0.0945	0.4599	0.2115***
To. Hb, g	2845.57	- 3.8286	0.0224	0.00050 ^{ns}
To. Hb, g / Kg. Bw	6.279	0.0399	0.2741	0.0751***
Aldosterone, pg/ ml	34.660	- 0.5234	0.8455	0.7149***
Cortisol, ng/ ml	7.194	0.1123	0.8051	0.6483***
TBF, L%	54.084	0.4153	0.6149	0.3781***
ECF, L %	13.810	0.3607	0.7066	0.4993***
ICF, L %	40.274	0.0546	0.1543	0.0238*
ISF, L%	11.375	0.2947	0.6611	0.4371***
PV, L%	2.434	0.0659	0.5462	0.2983***
BV, L%	4.057	0.0908	0.5319	0.2829***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, ns: not significant.

Equation in the form $Y = a + bx$, where $Y =$ any parameter study (dependent variable), $a =$ intercept of the characteristics, $b =$ regression coefficient for parameter on ambient temperature ($^{\circ}\text{c}$), $x =$ independent variable, $r =$ coefficient of correlation, $R^2 =$ coefficient of determination.

Ht%: haematocrite ratio; Hb, g%: hemoglobin content; To.Hb,g : total hemoglobin content; To.Hb, g/Kg.BW :total hemoglobin gm/ Kg live body weight; TBF, L%: total body fluids; ECF, L% extracellular fluid; ICF,L%: intracellular fluid; ISF, L%: interstitial fluid; PV, L%: plasma volume; BV, L%: blood volume.

Seasonal Effect on Body Fluids Compartment:

The absolute and relative values of total body fluid were greater significantly ($P < 0.05$ and $P < 0.01$) in summer than other seasons as shown in Table (5). The increase in TBF in hot climate may be an adaptive mechanism for heat tolerance, since it will allow the animal to store a great amount of heat because of the high specific heat of water during the hot part of the day and dissipate it during the cool hours (Yousef and Johnson, 1985). Moreover, Parker *et. al.*, (2004) in *Bos indicus* steers reported that the replacement of water from the gastrointestinal tract may have been responsible for maintenance of body water in stress.

The water pools in the ruminant body are dynamic, moving freely, from the lumen of the gastrointestinal tract to the extracellular fluid. This flux has resulted in considerable variation in the determination of body water loss from stressors (Cole, 1995).

Table (5): Effect of season on body fluids compartment in Buffaloes.

Parameters	Season			
	Summer	Autumn	Winter	Spring
TBF, L*	252.89± 14.709 ^a	243.18 ±12.545 ^{ab}	234.31 ±11.514 ^b	231.97 ±9.267 ^b
TBF,mL/kg**	691.07± 4.114 ^a	639.89 ±3.043 ^b	607.19 ±9.207 ^c	615.68 ±5.604 ^c
TBF,L%**	69.12 ± 0.411 ^a	63.99 ±0.304 ^b	60.72 ±0.921 ^c	61.57 ±0.560 ^c
ECF, L**	99.67 ± 7.028 ^a	84.79 ±5.650 ^b	77.75 ±4.850 ^b	79.21 ±4.986 ^b
ECF,mL/kg**	268.39± 2.631 ^a	220.13 ±2.398 ^b	198.56 ±1.663 ^c	203.71 ±1.720 ^c
ECF, L%**	26.84± 0.263 ^a	22.01 ±0.239 ^b	19.86 ±0.166 ^c	20.37 ±0.172 ^c
ICF, L	153.22± 7.853	158.39 ±7.054	156.56 ±6.798	152.76 ±4.713
ICF, mL/kg	422.67± 5.481	419.76 ± 3.751	408.63 ±5.876	411.97 ±10.213
ICF L%	42.27 ± 0.548	41.98 ±0.375	40.86 ±0.588	41.19 ±1.021
ISF, L*	83.07 ± 6.419 ^a	70.65 ±5.024 ^b	63.86 ±4.156 ^c	65.16 ±4.336 ^{bc}
ISF, mL/kg**	219.97± 3.617 ^a	181.80 ±2.535 ^b	162.74 ±1.588 ^c	166.84 ±1.803 ^c
ISF, L%**	21.99± 0.362 ^a	18.18 ±0.254 ^b	16.27 ±0.159 ^c	16.68 ±0.180 ^c

Average in the same row having different superscripts differ significantly * P < 0.05, ** P < 0.01. TBF: total body fluids; ECF: extracellular fluid; ICF: intracellular fluid; ISF: interstitial fluid.

Moreover Yousef and Johanson (1985) pointed out that high environmental temperature stimulates the drinking center in hypothalamus. This might contributed to increase the water turnover rate as adaptive mechanism. On the other hand, heat increased ADH secretion in cattle (El-Nouty 1980) this may be contributed to an increased in total body water. Also, El-Banna (1970) reported that the mechanism of water balance under hot conditions could be discussed on the basis of osmolarity of body fluids. The intravascular compartment is also affected since it was noted that the intravascular compartment constituents the immediate source of water loss under heat stress this decrease of plasma volume is associated with the increase in its colloidal and osmotic pressure (Mackfarlane, *et al.*, 1963) the decrease in plasma volume stimulates volume receptors in the carotid artery. These impulses are then transmitted to the hypothalamus, resulting in an increase in ADH secretion (El-Nouty 1980) and the volume receptors operate to maintain electrolyte balance. Furthermore, as environmental temperature increases the blood reaching hypothalamus, causes an increase in hypothalamic temperature (Carlson, 1962). This increase in hypothalamic temperature may stimulates other hypothalamic areas controlling water intake. It is also known that under the temporary conditions of water deficit, the thirst center is stimulated, and thus drinking of water is enhanced (Kamal, *et al.*, 1972). Also it seems likely that a high rate of water turnover during the evaporative cooling leads to a greater renal retention of water and electrolytes than is found in the steady state of unheated animals, where water is retained primarily, and adjustments of electrolytes follow. Moreover, El-Banna (1970) reported that buffaloes retained water more readily than Friesian cows.

The absolute values L and the relative ECF to body weight ml/kg or L% body weight was significantly increased (P < 0.01) in summer than in winter. Saxena and Joshi (1980), found no discernible change in TBF but there was significant increase in its ECF component with corresponding decrease in intracellular component when animals under 21-day stress at 37°C. They attributed the increase in ECF at 37°C mainly to the increase in serum volume.

The role of the ECF in homeostasis of internal body conditions (mainly osmosis and temperature) is very important and the comparison between the animals or breeds in seasonal changes of ECF must be based on its relative values to body weight (Ashour 1990). The relative volume of ECF increased greatly in summer compared to that in winter, this increase was furnished by excess of water intake in summer. The excess in ECF fulfill adequate supply of water for heat dissipation through water vaporization from the body surface during the hot summer (Ashour, 1990).

The extracellular regulating mechanisms appear to be a function of regulating extracellular fluid volume and composition; which depends essentially upon the regulation of these variables independently extracellular fluid concentration is regulated by control of free water excretion via regulation of ADH secretion, control of extracellular volume depends on the control of extracellular sodium, and is regulated by adrenal, renal and other mechanism, (El-Banna, 1970).

It appears from previous studies that ruminants living in hot environments, increased rate of water turnover which associated with enlargement of the plasma, and interstitial spaces. Moreover Bartter, (1963) suggested that extracellular volume is regulated through aldosterone action on sodium excretion and reabsorption in proximal tubules. However, in ruminants it seems that mineralocorticoid secretion falls, while the glucocorticoids secretion increases under heat stress, (Macfarlane, *et. al.*, 1963).

There was no significant difference between seasons with respect to intracellular fluid as absolute or relative values to body weight (Table 5). On the other hand the relative of ICF ml / kg.BW or L %. BW was greater in summer than in autumn, winter and spring. These results are in agreement with those obtained by El-Banna (1970), who found that the intracellular fluid (ICF) in buffaloes was increased as the result of the rise of temperature to 32°C, which caused an increase in the intracellular space (ICS) values but such increase was not statistically significant. Moreover, El-Banna (1970) reported that in buffaloes, the increase of water retention in the intracellular compartment at high temperature may be ascribed to excessive losses of sodium in sweat and urine, these results in a fall in extracellular osmolarity, followed by a shift of water to the ICS. The stability of volume and composition of the body fluids appears to be based upon the total intracellular water content when the fluid balance of the animal is subjected to sudden or extreme stress conditions. The volume of composition of the intracellular compartment is strongly defended against change at the expense of the extracellular compartment. In this connection, Mishra, *et. al.*, (1983) in buffaloes found that intracellular compartment is the last one to be affected by the high ambient temperature.

It is clear from Table (5) that the ISF absolute and relative to body weight increased markedly and significant ($P < 0.05$) and ($P < 0.01$), in summer than in autumn, spring and winter. Garg and Nangia, (1981) found that the changes in ISF volume may be attributed to its elasticity of the interstitial space which permits to absorb much of the fluctuating body water.

It was interesting to note that the significant positive relationships ($P < 0.01$ and $P < 0.05$) between ambient temperature and TBF, L %, ECF, L %, ICF, L % and ISF, L % (Table 4).

Seasonal Effects on Vascular Fluid:

The absolute values of PV and PV relative to body weight (ml/kg B.W or L % B.W) were greater significantly ($P < 0.01$) in summer than in autumn, winter and spring (Table 6). This biological increase of PV in summer is refer to the effect of

high environmental temperature on body temperature and supplying enough source of water for skin and respiratory vaporization (Ashour 1990). Macfarlane, *et. al.* (1963) stated that PV is to some extent maintained at the expense of ISF because the colloid osmotic pressure of plasma where plasma glucose and creatinin increased under hot condition (Chayabuter, *et. al.*, 1987). Proteins are the only osmotically active substances present at plasma ISF interface. The possible increase in colloidal osmotic pressure caused by increased plasma protein concentrations augments water passage from the extra vascular space to the intra vascular compartment (Ashour, 1990), Similar results were also reported by Ashour and Shafie, (1993) who found that in Egyptian buffaloes ISF and PV percentage were increased as response to heat stress solar radiation only was not paralld, inducing great increase in ISF. This arrangement of fluids in body compartment replenish more volume of ISF to insure permanente supply of fluid from this ISF to the plasma to maintain a stable blood volume against any water vaporization for cooling body temperature. Also the main factor causing the large increase of PV due to solar radiation effect was vasoconstriction of spleen expelling blood from its temporary store (Bass and Henschle, 1956).

Table (6): Effect of season on plasma and blood volume in Buffaloes.

Parameters	Season			
	Summer	Autumn	Winter	Spring
PV L **	16.30 ± 0.563 ^a	14.15 ± 0.643 ^b	13.89 ± 0.709 ^b	14.04 ± 0.662 ^b
PV mL/Kg **	48.27 ± 1.390 ^a	38.33 ± 0.662 ^b	35.82 ± 0.333 ^c	36.87 ± 0.430 ^c
PV L%	4.83 ± 0.139 ^a	3.83 ± 0.066 ^b	3.58 ± 0.033 ^c	3.69 ± 0.043 ^c
BV L	25.20 ± 0.940 ^a	22.51 ± 1.099 ^b	22.12 ± 1.197 ^b	21.69 ± 1.110 ^b
BV mL/Kg **	73.93 ± 1.921 ^a	60.37 ± 0.906 ^b	56.57 ± 0.518 ^c	56.57 ± 0.703 ^c
BV L%	7.39 ± 0.192 ^a	6.04 ± 0.091 ^b	5.66 ± 0.052 ^c	5.66 ± 0.070 ^c

Average in the same row having different superscripts differ significantly * p < 0.05

** p < 0.01. PV: plasma volume; BV: blood volume.

Farrell and Taylor (1962) stated that under condition of electrolyte balance, the osmoreceptors mechanism serves the function of maintaining fluid volume. If the total body electrolytes remains constant, changes in osmolarity reflects changes in body water. Thus when the mechanisms of maintaining electrolyte balance, functions properly, the osmoreceptors workers in accordance with volume receptors. In case of persistence negative sodium balance, the osmoreceptors mechanism fails to functions as protective device for blood volume. Under these circumstances there should be a sacrifice in volume for osmolarity in case of critical reduced volume, the volume receptor mechanism is prepotent and induces ADH secretion despite the resultant dilution of plasma electrolytes.

There was significant increase (P<0.01) in each of absolute and relative values of BV (L% and ml/kg. BW) under hot conditions, in summer than in others seasons (Table 6).

Ashour and Shafie (1993) found that solar radiation exposure caused rapid movement of minerals together with water to the extracellular fluid (ECF) to create further increase of interstitial fluid volume (ISF) and maintain plasma volume (PV) with normal osmolarity; this transfer may surpass the loss in BV by vaporization.

Kamal, *et. al.*, (1993) also found similar results in non-pregnant non-lactating water buffaloes. He found that BV ml/kg BW of water buffaloes increased by + 9.07 % in summer these increased of vascular fluids was transfer more heat from body core to the skin to be dissipated by evaporative and non evaporative cooling mechanisms.

True enough there was positive relation ($P < 0.01$) between vascular fluids and ambient temperature ($^{\circ}\text{C}$) as shown in Table (4).

Body and vascular fluids are essential for both controlling of homeostasis and heat balance. Therefore, seasonal variations present in thermoregulatory parameters, blood serum constituents, body pools of minerals, aldosterone and cortisol along with body and vascular fluids in non pregnant non lactating buffaloes under Egyptian climatic conditions. It could be concluded that year seasons have an effect on the physiological parameters under study and we can predict these parameters under any ambient temperature in Fayoum province environment.

REFERENCES

- Ashour, G. (1990). Water balance in Bovine as related to heat regulation. Ph.D. Thesis. Fac. of Agric. Cairo. Universty. Egypt.
- Ashour, G. and M.M. Shafie, (1993). Water balance in riverine buffaloes.1- Mobilization of body fluids under heat and dehydration stresses. Proc. Inter. Symp, Cairo, Egypt, 9-12-Nov., EAAP Pub.No.62: 194.
- Ashour, G. (1993). Stress response of blood volume and haematocrit in Egyptian buffaloes. Proc. Inter. Symp., Cairo. Egypt, 9-12 Nov., EAAP Pub. No. 62:191.
- Ashour, G; M.M. Shafie and H.M. Morad (2001). The significance of rumen fluid volume in sodium and potassium balance in buffaloes. Egyptian. J. Nutrition and Feeds: 4 (Special Issue) 473 484. Proc. Canf.Anim.Nutrition. Sharm El-Sheik, 23-26 Oct.
- Bartter, F.C. (1963). Regulation of the volume and composition of the extracellular and intracellular fluid. Ann.N.Y. Acad.Sci. 110. II: 682.
- Bass,D.E. and A.Henschle, (1956). Response of body fluids compartment to heat and cold. Physio. Rev. 36: 128
- Beltagy, H.G.M., (1990). Effect of some environmental and heamatological factors on blood biochemical and heamatological characteristics of pure-breed Friesian cows. M.Sc. Thesis, Alexandria Univ., Egypt.
- Beng,C.G. and K.L. Lim (1973). Colorimetric determination of albumin in serum. Amer. J.Clin.Path.,59:14.
- Bligh, J. (1975). The initial of thermal polypnoea in calf. J. physiol. 136 : 413.
- Brodie, B.B.; J. Axelord; R. Soberman, and B. Levy, (1949). The estimation of Antipyrine in biological materials. J.Biol. Chem., 179: 25.
- Burtis, A., (1999). Text Book of clinical chemistry. 3rd Ed, AACC,Press.
- Cannon,D.C., (1974). Assesment of total protein in clinical chemistry-principles and techniques. 2nd Ed. RJ Henry,et.al.,Eds. Harper and Row, Hagerstown, MD, 411.
- Cardozo, R.H., and I.S. Edelman (1952). The volume distribution of sodium Thiosulfate as a measure of the extracellular fluid space. J. Clin. Invest. 31, 3: 280.
- Carlson, L.D. (1962). Temperature. Annu. Rev. Physiol. 24: 85-108.

- Chaiyabutr, N.; C.Buranakarl; V. Muangcharoen; P.Loypetjra and A. Pichaicharnarong., (1987). Effect of acute heat stress on changes in the rate of liquid flow from the rumen and turnover of body water of Swamp buffalo. *J. Agric. Sci. Camb.*, 108: 549.
- Cole, N.A. (1995). Influence of a three – day feed water deprivation period on gut fill, tissue weights, and tissue composition in mature wethers. *J. Anim. Sci.* 73: 2548.
- Davis, H.A.; and L. Isenberg, (1953). An improved blood volume method (Evan's blue dye) utilizable even in the presence of hemolysis and or lipemia. *J. Lab. and Clin.Med.*41: 789.
- Duncan, (1955). Multiple ranges and multiple F test. *Biometric*, 11: 1.
- El-Banna, I.M. (1970). The effect of high environmental temperature on body fluid compartments and water metabolism in farm animals. M.Sc. Thesis, Fac. Of Agric. Ain – Shams Univ. Cairo, Egypt.
- El-Masry, K.A.; H.A.El-Foulyand, and S.A. Gabr., (2001). Physiological action of chromium on some blood biochemical constituents and immune response in relation to growth performance of heat stressed calves. *Egyptian, J. Nutrition and Feeds.* 4: 453.
- El-Nouty, F.D.; I.M. El-Banna, and H.D. Johnson (1978). Effect of adrenocorticotrophic hormone on plasma glucocorticoids and antidiuretic hormone of cattle exposed to 20and 30 C°. *J. Dairy Science*, 61: 189.
- El-Nouty, F.D.; I.M. El-Banna; I.P. Davis, and H.D. Johnson, (1980). Aldosterone and ADH response to heat and dehydration in cattle. *J. Appl. Physiol.*, 48: 249.
- Farrell, G; and A.N.Taylor., (1962). Neuroendocrine aspects of blood volume regulation. *Annu. Rev. Physiol.* 24:271.
- Findlay, J.D.and D.L.Ingram, (1961). Brain temperature as a factor in the control of thermal polypnoea in the OX (*Bos Taurus*). *J. physiol.* 155: 72.
- Garg, S.K. and O.P. Nangia., (1981). Response of body fluid compartments to climatic variations in buffaloes. *Indian. J. Anim.Sci.*, 51: 1022.
- Habeeb,A.A.M.; A.E.B. Zeidan, and A.A. Shitta, (2001). Role of Niacin and sprinkling in improving milk yield, composition and biochemical functions of the heat stressed Friesian cows. *Egyptian J. Nutrition and Feeds.* 4: 285.
- Hamed, Madeha, H.A.; M.S. Abd El- Fattah; H.A. Gawish, and M.T.A. Badawy, (2007). changes in body fluids and water balance of desert sheep and goats during water restriction. *Egypt. J. Basic Appl. Physiol.*, 2: 361.
- Hassan, G.A. and El- Nouty, F.D., (1985). Thyroidal activity in relation to reproductive states seasonal variations and milk production in buffalo and cow heifers during their first lactation. *Ind. J. Dairy Sci*, 38: 129.
- Kamal T.H.; M.K. Shebaita, and I.I. Ibrahim, (1993). Seasonal variations in different Physiological parameters in water buffaloes and Friesians. *Proc. Inter. Symp.*, Cairo .Egypt, 9-12 Nov., EAAP Pub. No 62:201.
- Kamal,T.H.; M.K. Shebaita, and I.M. El-Banna, (1972). Effect of heat and water restriction on water metabolism and body fluid compartments in farm animals. *Proc.Series, IAEA, Vienna*, 95.
- Kreiger, D.T. (1975). Rhythms of ACTH and corticosteroid secretion in health and disease and their experimental modification. *J. Steroid Biochem* 6: 785.
- Macfarlane, W.V.; R.J.H. Morris; B. Howard (1963). Turnover and distribution of water in desert camels, sheep, cattle and Kangaroos. *Nature*, 197: 270.

- Makaren, A. (1974). Hemoglobins, myoglobins and haptoglobins. In: Clinical Chemistry. 2nd Ed., Edited by Henry, R.J., D.C. Cannon and J.W. Winkelman, Harper & Row, Publishers, Hagerstown, Maryland, New York, USA, 1131.
- Marai, F.M.; A.H. Daader,; A.M. Abdel-Samee, and H. Ibrahim, (1997). Winter and summer effects and their amelioration on lactating Friesian and Holstein cows maintained under Egyptian conditions. International Conference on Animal Production and Health, 305.
- Mayes, D. (1970). Radioimmunoassay for plasma aldosterone. J. Clin. Endocrinol. Metab, 30 : 682.
- Mclean, J.A. (1963). The partition of sensible body weight and heat stress from cattle under various climatic conditions. J.physiol. 167: 424.
- Migeon, C.J. and R.L. Lanes, (1990). Adrenal cortex : hypo and hyper function. IN Lifshitz F(Ed): Pediatric Endocrinology, A clinical Guide, Second Edition. Marcel Dekker, INC., New Yourk, 333.
- Mishra, U.K.; S.K. Saxena; H.K.B. Parekh; V.K.Guru; I.J. Sharma and H.S. Singh, (1983). Effect of season on body water distribution in three genotypes of crossbred calves. Indian.J. Dairy. Sci. , 36: 21.
- Mudron, R; J. REHAGE; H.P. Sallmann; M.Höltershinken and H. Sochlz., (2005). Stress response in dairy cows related to blood glucose. ACTA Vet. Brno,74: 37.
- NRC (1988). Nutrient Requirements of Dairy cattle. National Research Council. 6th Revised Ed. Washington, D.C. USA.
- Pandey, M.D. and A. Roy, (1969). Studies on the adaptability of buffaloes to tropical climate: seasonal changes in the water and electrolytes status of buffalo cows. Indian. J. Anim. Sci., 39: 307.
- Parker, .A.J; G.P. Hamlin; G.J. Colmane and L.A. Fitzpartick., (2004). Excess cortisol interferes with a principal mechanism of resistance to dehydration in *Bos Indicus* Steers. J. Anim. Sci. 82: 1037.
- Reece, W.O., (1991). Physiology of domestic animals. Lea and Febiger. Philadelphia.London.
- Reitman,S. and S. Frankel, (1957). A clorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases J. Clin. Path., 28: 56.
- Saxena, S.K. and B.C. Joshi, (1980). Dynamics of body water under environmental heat stress. Indian. J.Anim.Sci.,50: 383.
- Shebaita, M.K. (1993). Indices of acclimatization, acclimation or solar radiation, they all are the same.7th Conf. Anim. Prod. Edmonton, Canada, 293.
- SPSS (1997). Statistical package for social science. Release 8.0, copyright © SPSS. INC., Chicago, U.S.A.
- Sunderman, F.W. Jr. and F.W. Sunderman (1958). A turbidimetric method for potassium determination. Am. J. Clin. Pathol. 29:55.
- Trinder, P., (1951). A colorimetric method for sodium determination. Analyst, 76: 596.
- Yousef, M.K. and Johnson, H.D., (1985). Stress Physiology in livestock. Vol. I, CRC Press, Inc, Florida.

تأثير الاختلافات الموسمية على سوائل الجسم والاعوية الدموية
في الجاموس تحت ظروف المناخ المصري

منى عبد التواب الخشاب ، على ربيع عبدالرحمن و عمر محمد حيدر
قسم الانتاج الحيوانى - كلية الزراعة - جامعة الفيوم - الفيوم - مصر .

استخدم في هذه الدراسة ١٢ انثى غير حلابة وغير حامل من الجاموس المصرى فى مزرعة كلية الزراعة - جامعة الفيوم. عمر الحيوانات يتراوح من ٤ : ٥ سنوات وكانت تعيش تحت مظلات الحديد المجلفن طول العام (يونيو ٢٠٠٤ : مايو ٢٠٠٥) لدراسة تأثير مواسم السنة على كل من مقاييس التحمل الحرارى مثل درجة حرارة الجسم، التنفس ومعدل النبض كذلك تم قياس كل من الهيماتوكريت والهيموجلوبولين.

تم فصل عينة من سيرم الدم لتقدير كل من البروتين والاليومين والجلوبيولين كذاك تم تقدير انزيمات الكبد، الصوديوم، البوتاسيوم والكلوريد كما تم قياس ماء الجسم الكلى والماء داخل وخارج وبين الخلايا ايضاً مع قياس حجم البلازما والدم بالجسم.

أوضحت النتائج اختلافات معنوية لتأثير موسم السنة فى كل من معدل التنفس والنبض وكذلك زيادة مستوى البروتين الكلى والاليومين والجلوبيولين فى الشتاء عن الصيف. كما وجد زيادة نسبة هرمون الالدوستيرون فى الشتاء عن الصيف كما اوضحت نتائج تحليل الكوريتزول عكس هذا.

أدى ارتفاع درجة حرارة الصيف الى زيادة فى سوائل الجسم سواء ماء الجسم الكلى - الماء داخل الخلايا وماء خارج وبين الخلايا ايضاً زيادة فى حجم الدم والبلازما كنوع من اقلمة الحيوان للجو الحار فى موسم الصيف عنه فى باقى المواسم.