

Egyptian Journal of Veterinary Sciences

https://ejvs.journals.ekb.eg/



Impact of Reproductive Status, Body Condition Score, and Locality on Hormonal, and Some Blood Metabolites in Egyptian Buffaloes

Seham S. Soliman^{*}, Amro M. El-Sanea^{*}, Omaima M. Kandil, Amal M. Aboelmaaty and

Ahmed Sabry S. Abdoon

Department of Animal Reproduction and Artificial Insemination, Veterinary Research Institute, National Research Centre, Dokki 12622, Cairo, Egypt.

Abstract

HIS STUDY was designed to find out the influence of reproductive status, body condition, and localities on hormonal profile, antioxidant status, trace elements, blood biochemical, and _ metabolites in Egyptian buffaloes. Three experiments were conducted, in experiment 1; buffalo were divided into two groups according to localities (Fayoum & Behira). In experiment 2, 97 buffalo were divided into cyclic (n= 29), early pregnant (n=43), inactive ovaries (n= 12), and endometritis (n= 13). In experiment 3, according to body condition score, buffaloes were divided into three groups (Low, medium, and high). Blood samples were collected from all animals for analyzing progesterone (P_4), estrogen (E_2), superoxide dismutase (SOD), malondialdehyde (MDA), total protein (TP), albumin (Alb), total cholesterol, triglycerides, low- density lipoprotein (LDL), glucose, Zinc (Zn), and copper (Cu). Results indicated that P4, E2, antioxidant, TP, Alb, Glob, A/G ratio, glucose, Zn and Cu levels significantly increased (P<0.05) for buffaloes raised at Behira (Delta) compared to Fayoum (Upper Egypt). In pregnant and cyclic buffalo, P4, E2, antioxidant, TP, Glob, A/G ratio, cholesterol, triglycerides, glucose, SOD, Zn and Cu increased (P<0.05) in comparison to those with inactive ovaries or endometritis. Moreover, buffaloes with high and medium body conditions had higher (P < 0.05) levels of P₄, E₂, TP, Alb, glob, total cholesterol, triglycerides, and Cu than those having low body condition score. MDA and LDL levels were high (P<0.05) in animals with low body condition score. In conclusion, blood serum concentrations of P4, E2, total protein, albumin, globulin, total cholesterol, LDL, triglycerides, Zn, and Cu were the most sensitive biochemical markers to represent the reproductive status of the animals. The body condition score and locality have impacted the fertility of dairy buffalo.

Keywords: Buffalo, Locality, Reproductive status, Body condition score, Hormone, Biochemical analysis.

Introduction

In Egypt, for the annual production of milk and meat, cattle and buffaloes together account for 70% of all animal production [1]. From 3,250,000 heads in 1993 to 4,200,000 heads in 2013, the buffalo population rose [2]. Three years later, the buffaloes number decreased to be 3.5 million head during 2016 [3]. Buffalo is suffering from low fertility such as delayed puberty, long inter-calving intervals, seasonality of breeding and poor estrus expression. In addition, many problems may limit their productivity such as poor nutrition, climatic changes, management and genetic parameter [4]. One of the most critical factors affecting buffalo productivity is

the weather. Environmental stress causes sequences of variation in the biological functions of animals. This includes oxidative stress, a decrease in food intake, changes in blood biochemistry and hormonal [5]. Thereby reducing the assembly performance of the animals, it is the primary indicator of the animal's physiological and pathological conditions [6]. The high humidity and relatively high temperature are less favourable for reproduction in buffalo than in the winter season. It has been observed that the conception rate and manifestation of estrus behaviours are low throughout June to August [7]. As a result of increased reactive oxygen species (ROS) levels caused by heat stress, antioxidant enzymes such as glutathione peroxidase, catalase and superoxide dismutase increase [8]. In buffalo,

*Corresponding author: Samir Soliman E-mail: sehamsamir81@yahoo.com. Tel: 00201273052419 (Received 05/12/2023, accepted 31/01/2024) DOI: 10.21608/EJVS.2024.252235.1699

^{©2024} National Information and Documentation Center (NIDOC)

progesterone concentrations are influenced by both environmental and nutritional status, and low progesterone concentrations are associated with high embryonic mortality, low oocyte vigor, and slow endometrium growth [9]. Furthermore, the body condition score (BCS) is a simple and reliable way of measuring the animals' nutritional and health status. The BCS is a quick and easy approach for estimating an animal's energy reserve [10], and it is thus related to energy balance status and fertility [11]. Energy deficiencies delay the restart of postpartum estrous cycles and diminish the first insemination conception rate, resulting in low fertility [12]. Since BCS is related to dairy cow fertility, the occurrence of illnesses [13]. Understanding the metabolic physiological factors affecting the reproductive potentials of buffalo will open new gates for improving their productivity and fertility. Therefore, this work was designed to evaluate the effect of geographical areas (Locality in one governorate in the North Nile Delta) and in another governorate at the beginning of Upper Egypt), reproductive status, and BCS on hormonal profile, antioxidant status, trace elements, blood biochemical and metabolites parameters in Egyptian buffalo.

Material and Methods

Ethical approval

This experiment was approved by the Institutional Medical Research Ethical Committee, National Research Centre, Egypt (Project ID: RO120201). The work was conducted under the provision of relevant Egyptian laws, and Good Medical and Laboratory Practice (GCP and GLP) guidelines. The experimental work was conducted during the period from September 2020 to June 2022.

Experimental design

Experiment 1: Effect of Localities on hormonal, antioxidant, trace elements, blood biochemical, and metabolites changes in Egyptian Buffaloes.

Field visits were organized in collaboration with the Department of Reproductive Management belongs to the General Organization of Egyptian Veterinary Services (GOVES). Two visits per month were performed to different villages at Fayoum Governorate (Upper Egypt, n=63), and Beheira (Lower Egypt, n=90). During the visit day, the temperature and humidity were recorded. At Behira the average temperature was 25±5°C. The maximum temperature-humidity index (THI) was 75%. At Fayoum governorate, the average temperature was 34±3°C, and the maximum THI was 78%. At both Fayoum and Behira buffaloes were gynecologically examined to determine their reproductive status. According to the case history, rectal palpation, and ultrasonography findings, animals were classified into animals with smooth inactive ovaries (BLSO; n=79), animals suffering from endometritis

(Endomet; n=91), early pregnant animals (Preg; n=24), and cyclic animals with dominant follicles and/or regressed corpus luteum or with mature CL (Cyclic; n=51).

Experiment 2: Effect of reproductive status on hormonal, antioxidant, trace elements, blood biochemical, and metabolites changes in Egyptian buffaloes.

During winter and summer seasons, a total of 263 buffaloes raised at Behira Governorate were used in this study. Animals were gynecologically examined to determine their reproductive status. According to palpation, history. rectal the case and ultrasonographic findings, animals were classified into smooth inactive ovaries (BLSO, n=79), animals suffering from endometritis (n=91), early pregnant animals (n=42), and cyclic animals with dominant follicles and regressed corpus luteum or with mature CL (cyclic animals, n=51).

Experiment 3: Effect of body condition scores on hormonal, antioxidant, trace elements, blood biochemical, and metabolites changes in Egyptian buffaloes

A Total of 124 buffalo in the Fayoum governorate were used in this experiment. The BCS was evaluated using the following score approach given by Yunober Mberato et al. [14] score 2.0-2.5 - low; with observed round transverse processes and little fat covering during palpation (n=72); score 2.6-3.0 -medium (n=38); with slightly unfelt transverse processes as a result of good muscular and thick fat covering during rigid pressure; score 3.1-3.5 –high (n=23).

Blood sampling

In all experiments, following gynecological examination, blood samples were taken at 10 to 11.0 a.m. from all animals under investigation using vacutainer tubes without EDTA for hormone and blood biochemical analysis. The blood samples were immediately placed on ice and kept at 4 °C for at least 2 hours. The samples were then centrifuged at 3000g for 30 minutes at 4 °C to obtain the serum, which was subsequently frozen at -20 °C.

Hormonal analysis

 P_4 and (E17- ß) levels in serum were determined using an Enzyme Linked Immuno-Sorbant Assay (ELIZA) kit (DE-EIA-STEROID-PROGESTRONE, DSI S.r.I Company, Italy) and Cat. No. BC-111 from BioCheck, Inc, Foster City). and the assay minimal detection limit was 0.2 ng /ml for p4. The variation intra assay coefficients for low and high references were 2.30% and 4.5%.

Serum protein analysis

Total protein and albumin levels were determined using a colorimetric method with a specialized kit (Biodigonstic Company, Giza, Egypt). Serum globulin concentration was calculated by subtracting albumin concentration from the total protein content in the same sample. The A/G ratio was calculated by dividing the albumin concentration by the globulin concentration.

Blood metabolites

Enzymatic colorimetric kits (Biodiagnostic Company, Giza, Egypt) were used to determine triglycerides, total cholesterol, and LDL levels.

Blood antioxidants

The amounts of SOD and MDA were determined using a colorimetric method using a particular kit (Biodiagnostic Company, Giza, Egypt).

Trace elements

The levels of zinc and copper were determined using a colorimetric approach using a specific kit (Biodiagnostic Company, Giza, Egypt).

Statistical analysis

The SPSS statistic package (SPSS for Windows 20.0, SPSS Inc., Munich, Germany) was used for descriptive statistical analyses. A one-way ANOVA with different animal groups and an independent sample t-test were performed. The results are shown as means with standard error of the mean (SEM). The level of significance was fixed at < 0.05.

Results

Gynecological examination (Table 1) revealed that the number of RB buffaloes was higher (P<0.05) and the number of pregnant and cyclic buffaloes was lower (P<0.05) at Fayoum compared with Behaira governorate. The incidence of BLSO was similar between Fayoum and Behaira governorate. The effect of locality (THI) on hormonal, antioxidant, trace elements, blood biochemical, and metabolites is demonstrated in Table 2. Results indicated that the percentage of P₄, E₂, total protein, albumin, globulin, (A/G) ratio, total cholesterol, triglycerides, glucose, zinc, and copper levels were significantly (P<0.05) higher for buffalo raised at Behira compared to that in Favoum. On the other hand, buffaloes at Favoum have high (P<0.05) concentrations of LDL, MDA and (SOD). Buffaloes expressed a higher (P<0.05) incidence of BLSO during the summer season than the winter season. Also, the percentage of endometritis was higher (P<0.05) in winter than in the summer season. While, the percentage of pregnant animals and cyclic buffaloes did not vary between summer and winter season, however, they are significantly lower than BLSO and endometritis in both summer and winter seasons (Table 3). The effect of reproductive status on hormonal, antioxidant, trace elements, blood biochemical, and metabolite levels in buffalo is illustrated in Table 4. The results showed that the mean serum levels of estradiol, total protein, albumin, globulin, A/G ratio

and cupper were significantly high in cyclic buffalo than in other reproductive statuses, and the mean serum levels of triglycerides, total cholesterol, glucose, SOD and zinc were significantly (P<0.05) higher in cyclic and early pregnant buffalo than in other reproductive statuses. The mean serum levels of P₄, E₂, total protein, albumin, globulin, A/G ratio, triglycerides, total cholesterol, glucose, SOD, zinc and copper in inactive ovaries and endometritis were significantly (P < 0.05) lower than in other reproductive statuses in buffalo. The levels of MDA and LDL were significantly (P < 0.05) higher in inactive ovaries and endometritis animals than in cyclic, and early pregnancy.

The effect of BCS on hormonal, antioxidant, trace elements, blood biochemical, and metabolite levels in buffalo is presented in Table 5. The results showed that the mean serum levels of P₄, E₂, total protein, albumin, globulin, triglycerides, and total cholesterol and cupper were significantly (P < 0.05) higher in high and medium BCS than buffalo with low body condition scores. However, there were no significant differences in mean serum glucose levels among different BCS. The mean serum levels of SOD and LDL were significantly (P < 0.05) lower in low BCS than in high and medium body condition scores. In animals with high BCS, the mean serum levels of Zn, SOD, A/G ratio were significantly (P <0.05) higher than other BCS. In buffalo with medium BCS, the mean serum levels of SOD were significantly (P < 0.05) higher than low BCS.

Discussion

The current study found that the number of RB buffaloes was higher (P<0.05) and the number of pregnant and cyclic buffaloes was lower (P<0.05) at Fayoum (high THI) compared with Behira Govornerate (low THI). These findings are consistent with previous results Faraz et al. [15] who reported that high environmental temperature reduces feed intake, which leads to poor estrus expression and increased ovarian inactivity. The higher incidence of reproductive problems in buffalo in the hot localities may be attributed to the poor adaptability of the buffalo to the hot climates and more adverse conditions in hot areas.

In addition, our result reported that P_4 , estradiol, total protein, albumin, globulin, albumin/globulin ratio, total cholesterol, triglycerides, glucose, zinc and copper levels were higher in cold areas (Behira) than in hot areas (Fayoum). These results are consistent with [16, 17]. The corpus luteum functions are reflected in the P4 level, and any variation in this level may have profound physiological effects on reproduction. These results agree with [18-20] claimed that cooling the microenvironment of animals could improve serum total protein, globulin, and albumin content. Abdel-Ghani *et al.* [21] the higher level of glucose may be the result of protein's beneficial effects on animal nutrition, which raises the blood glucose levels of animals. In contrast, Costanzo et al. [22] reported that the environmental temperature did not affect the level of these parameters. Xulu et al. [23] discovered that the dry area had greater total protein levels, cattle might dehydrate during the cold temperature which may increase the concentration of the plasma proteins. Decreased level of serum total protein at Fayoum (high temperature) opposed that heat-stressed animals may be in the stage of hemodilution. Pandey et al. [24] reported that the high temperature period, the serum cholesterol level was tended to lower thyroid activity which decreased the feed intake during the hot environment and consequently reduced the intake of dietary cholesterol.

The mean levels of LDL, SOD and MDA in the hot weather locality (Fayoum) were significantly (P < 0.05) higher than in the cold weather locality (Behira). These results are in agreement with those reported by Sakatani et al. [25] and Abdoon et al. [26]. However, these results are contrasted with those reported by Lallawmkimi et al. [27]. SOD is considered one of the main molecules of intracellular antioxidant defense and it is obligatory in efficient defensive systems [28]. Therefore, measuring these markers is very important to evaluate the effect of high temperature locality on the oxidative stress levels of buffalo. High MDA levels occur due to high thermal temperatures. One of the main causes of oxidative stress, which results from a decline in antioxidant defense and an increase in the formation of free radicals and has anti-gonadotropic and antisteroidogenic effects, may be the increased lipid peroxidation found in the hot areas [29].

The current investigation found that serum progesterone levels in buffalo were considerably (P<0.05) higher in early pregnant animals than in cyclic, with the lowest concentrations seen in smooth inactive ovaries and endometritis and mean serum levels of estradiol were significantly lower in inactive ovaries than in cyclic buffalo. These results agree with that reported by Abdoon et al. [17]. The serum estradiol levels provide further evidence that serum levels are always a good index of follicular status. Minor changes in P4 concentration can influence embryo development, as experimentally induced P₄ concentration reductions altered follicular steroidogenesis and enhanced uterine PGF2a production [30]. It has been claimed that extreme environmental conditions along with under nutrition may be the cause of buffalo's protracted anestrous phases, in addition to delayed ovulation causing changes in the preovulatory follicle microenvironment reduced levels of steroids may be due to a decrease in the availability of the cholesterol precursor [31]. In this regard, data obtained from the present study implicated significantly (P 0.05) lower values of serum total protein, albumin, globulin, total

cholesterol, triglycerides, zinc and copper in the animals affected with smooth inactive ovaries and endometritis when compared to other groups. Our results are in agreement with that reported by Abd El-Razek and Allam [32], Hafez [33]. The antioxidant activity of element contracted the levels of both apoptosis and autophagy in the level mature oocytes. El-Sheikh et al. [34] and Mondal and Paul [35] reported a significantly higher value of total serum protein in cyclic than repeat breeder and inactive buffaloes that could be brought on by a lack of specific amino acids needed for the production of gonadotropins and gonadal hormones, which would result in reproductive hormonal abnormalities in animals and inactive ovaries [32]. Reproductive problems were thought to come from changes in the biochemical components of the blood rather than being a cause of them [36]. It is possible that globulin, acting as a copper transporter protein, affected the production of particular coenzymes, and hence the steroidogenesis during the early luteal phase of the cycle [35]. These results disagree with those reported by Ahmad et al. [37], Damptey et al. [38]. This high albumin level in regularly cycling cows indicated an increased requirement for amino acids and protein to produce GnRH and LH to trigger ovulation. In the current study, a lower concentration of total blood protein, albumin and globulin was associated with lower fertility. Feeding with low protein content can reduce fertility and decrease the number of services per conception. The current study suggests that the reproductive status of buffaloes influences total protein, albumin, and globulin concentration. Also, the present study advocates that serum cholesterol was the factor responsible for the exhibition of differential fertility. The amount of steroid hormones secreted depends on the availability of serum cholesterol in the microcirculation, which serves as a precursor for the production of steroid hormones. Therefore, a drop in the cholesterol level in the buffaloes with inactive ovaries and endometritis would be the reason why inactive buffalo are so common in the herd.

Reactive oxygen species (ROS), which are associated with the control of numerous reproductive processes, are subject to defense by superoxide dismutase, an important antioxidant. Superoxide dismutase and glucose levels were greater in samples from cyclic pregnant buffaloes. These results are in agreement with that reported in buffaloes by Abdoon et al. [39]. However, these results are contrasted by those reported in cattle Mondal and Paul [35]. The results of the present study may indicate that glucose levels may affect ovarian activity. According to studies, serum glucose plays a significant role in regulating reproduction, and when it is present at lower levels, it is thought to be the root of noncyclicity and decreased fertility rates. Significantly higher MDA levels in smooth inactive and endometrities than cyclic and pregnant buffaloes.

1391

These results are in agreement with that reported by Abdoon et al. [26]. Abd El-Razek and Allam [32] claimed that buffalo cows with inactive ovaries had reduced fertility and had elevated MDA readings. It is generally known how crucial copper is for growth, production, and reproductive processes. Ceruloplasmin and superoxide dismutase are two examples of proteins that contain copper and are crucial for copper's physiological processes El Wishy [40]. According to a study, Cu deficiency may harm female reproduction by causing the hypothalamuspituitary-gonadal axis to secrete less LH, which reduces ovarian estradiol secretion and prevents animals from expressing estrus [41]. Given these results, it is tempting to hypothesis that even a little drop in the serum Zn and Cu levels in buffalo could result in issues including aberrant ovarian development, disruption of the estrus cycle, and decreased FSH and LH synthesis/secretion, which can result in reproductive illnesses.

The mean serum levels of estradiol and progesterone, total protein, albumin, globulin, and A/G ratios triglycerides, total cholesterol, LDL, zinc and copper with high and medium body condition scores were significantly (P < 0.05) higher than buffalo with low body condition scores. our findings agree with those reported by Sirotkin et al. [42], Thembinkosi et al. [10] and Mouffok et al. [43], and with those of Manzoor et al. [44] who discovered that granulosa cells isolated from the ovaries of BCS2 cows secreted more estradiol than those isolated from the BCS3 group. Emaciation is connected with a decrease in ovarian follicular development and an increase in ovarian estrogen release. Yaylak et al. [45] reported that mean levels of triglycerides, cholesterol and albumin increased in high condition scores cows. On the other hand, also, our results were contrasted by that reported by Mouffok et al. [43] who found that the level of triglyceride in serum is greater in thin cows. Sirotkin et al. [42] found that cows with a tendency towards emaciation (BCS2) had significantly higher concentrations of Zn^{2+} in blood plasma than average body condition score (BCS3). Aktas et al. [46] reported that when compared to cows with thinner BCS, cows with a medium BCS tended to have lower serum LDL concentrations. Protracted duration of postpartum anestrus and decreased fertility are the results of negative energy balance, which reduces LH pulse frequency and decreases blood sugar, insulin, and insulin-like growth factor levels Manzoor et al. [44]

There were no significant differences among mean levels of glucose with different body condition scores. These results are in agreement with those reported in cattle by Lake *et al.* [47]. On the other hand, our results were contrasted with that reported in cattle by Aktas *et al.* [46]. Moreover, the results of the present study showed that there were significant

differences among mean serum levels of SOD and MDA with different body condition scores. These results are in agreement with that reported by Derkx [48]. The disparity could be attributed to differences in grades on the BCS scale, changes in physiological conditions, regional eating customs, race, and sex [39] revealed that the cold environmental conditions and the reproductive status have important effects on fertility in cattle and buffalo. The extraordinary adaptation mechanisms of buffalo can be used to explain changes in feed availability as well as variations in blood biochemical levels. Additionally, the relationship between feed quality and quantity and buffalo physiological and health state could be ascertained using the mean score values of blood biochemical markers based on animal status and location. Generally, the results of the current study showed that locale, reproductive state, and body condition score all significantly affected the nutritional status and blood metabolites of buffalo.

Conclusions

Low humidity and temperature locality are ideal for preserving physiological levels of hormones and metabolic factors, which may improve the buffalo's reproductive performance. The selection of climateresilient animals is necessary to secure the future of sustainable animal production. The reproductive status variations in biochemical profile suggest a need to consider these factors when choosing fertile animals because they can influence nutritional status, health status, and reproductive performance. Serum hormonal, antioxidant, trace elements, blood biochemical, and metabolites of Egyptian buffalo varied with, locality, reproductive status, and body condition score. A decline in blood parameters in different body condition score suggests a need for supplementary feeding

Conflict of interest

The authors declare that they have no conflict of interest.

Acknowledgment

Not applicable

Ethics approval and consent to participate

The experiments were performed in accordance with the recommendations of the National Institute of Health Guide for Care and Use of Laboratory Animals (publication no. 85-23, revised 1985). The experimental protocol for the in vivo experiments was approved by the Institutional Animal Care and Use Committee of the National Research Centre of Egypt (Approval no: RO120201).

Funding statement

This research work was completely financed by the National Research Centre of Egypt Proj: RO120201.

Authors' Contributions

ASSA designed the experiment. ASSA, OMK, AME, SSS and AMA conducted the experimental work. ASSA drafted the manuscript. All authors were involved in scientific discussion, analysis of the data and revising the manuscript. All authors read and approved the final manuscript.

TABLE 1. Incidence of reproductive abnormalities in buffalo and cattle at Fayoum and Behaira

Governorate.

Locality		Reproductive status			
	BLSO	RB	Preg	Cyclic	
Fayoum	18(28.6)	38(60.3)*	3(4.8)	4(6.3)	
Behaira	24(30.4)	31(39.2)	16(20.3)*	19(24.1)*	

BLSO animals with bilateral smooth ovary; RB: repeat breeder; Preg: pregnant buffaloes; Cyc: Cyclic. *P<0.05

TABLE 2. Effect of localities (THI) on serum levels of some reproductive hormones, proteins, blood metabolites, blood antioxidants, and some trace elements in buffalo.

	THI of Governorate			
Parameter	High (Fayoum)	Low (El Behera)		
Progesterone (ng/ml)	0.38±0.02 ^b	1.2 ± 0.04 ^a		
Estradiol(pg/ml)	0.6±0.5 ^b	0.9 ±0.3 ^a		
Total protein (g\dl)	4.7 ± 0.1^{b}	7 ± 0.1^{a}		
Albumin (g\dl)	2.4±0.1 ^b	3.2±0.1 ^a		
Globulin (g\dl)	2.2±0.1 ^b	$4{\pm}0.1^{a}$		
Albumin/globulin ratio	0.6±0.04 ^b	0.9±0.4ª		
Triglycerides (mg/dl)	61.5±2.6 ^b	79 ± 2.9^{a}		
Total cholesterol (mg/dl)	97±5.1 ^b	117.8±4.3 ^a		
LDL (mg/dl)	111.5±5.7 ^a	55.4±3 ^b		
Glucose(mg/dl)	50.4±2.4 ^b	68.5±1.9 ^a		
SOD (u/ml)	109.5±6.2 ^a	78.6 ± 4.5^{b}		
MDA (nmol/ml)	11.7±0.4 ^a	8±0.1 ^b		
Zn (umol/l)	0.11 ± 0.001^{b}	0.13 ± 0.003^{a}		
Cu (umol/l)	0.25±0.01 ^b	0.32±0.01 ^a		

Superscripts with different letters within the same row differ significantly at P<0.05.

Season	No.				
	animals	BLSO	Endometritis	Pregnant	Cyclic
Summer	109	41 (37.6) ^b	26 (23.9) ^b	19 (17.4)	23 (21.1)
Winter	154	$38(24.7)^{a}$	65 (42.2) ^a	23 (14.9)	28 (18.2)

TABLE 3. Incidence of reproductive abnormalities in buffalo at Fayoum Governerate during Summer and winter seasons

a, b differ significantly within the same column at P<0.05.

TABLE 4. Effect of reproductive status on serum levels of some reproductive hormones, proteins, blood
metabolites, blood antioxidants, and some trace elements in buffalo (Mean ± SEM).

Parameters	Reproductive status			
	Cyclic	Inactive ovaries	Endometritis	Early pregnant
Progesterone (ng/ml)	0.7 ± 0.03^{b}	$0.24{\pm}0.02^{\circ}$	0.31±0.05 ^c	1.3±0.05 ^a
Estradiol(pg/ml)	$1.1{\pm}0.05^{a}$	$0.4{\pm}0.6^{c}$	$0.5{\pm}0.08^{\circ}$	0.8 ± 0.03^{b}
Total protein(g\dl)	$7.4{\pm}0.2^{a}$	3.6±0.2 ^c	3.9±0.3°	6.3±0.1 ^b
Albumin(g\dl)	3.4±0.1 ^a	1.9±0.2 ^c	2.3±0.2 ^c	2.9±0.1 ^b
Globulin(g\dl)	4.1 ± 0.2^{a}	1.8±0.2 ^c	1.8±0.2 ^c	$3.4{\pm}0.2^{b}$
Albumin/globulin ratio	1 ± 0.06^{a}	0.3±0.03 ^c	$0.4{\pm}0.02^{\circ}$	0.8 ± 0.04^{b}
Triglycerides(mg/dl)	83.3±4.1 ^a	55±2.1 ^b	56.2±4.9 ^b	72.2±3.2 ^a
Total cholesterol(mg/dl)	126±5.6 ^a	77±4.6 ^b	68.8 ± 4.5^{b}	117±4.4 ^a
LDL(mg/dl)	65.9±6 ^b	100.1±9.5 ^a	97±1.1ª	76.7±6.7 ^b
Glucose (mg/dl)	69.2 ± 2.4^{a}	43.5±3.6 ^b	44.8±3.5 ^b	62.7±1.9 ^a
SOD (u/ml)	97.5±7.8 ^a	59.7±1.1 ^b	66±1 ^b	96.6±6.5 ^a
MDA(nmol/ml)	8.3±0.3 ^b	11.8±0.8a	$11.2{\pm}0.8^{a}$	8.5 ± 0.4^{b}
Zn(umol/l)	$0.14{\pm}0.01^{a}$	0.11 ± 0.001^{b}	$0.10{\pm}0.001^{b}$	$0.14{\pm}0.006^{a}$
Cu(umol/l)	$0.4{\pm}0.03^{a}$	$0.18 \pm 0.008^{\circ}$	$0.18 \pm 0.008^{\circ}$	0.3 ± 0.01^{b}

Superscripts with different letters within the same row differ significantly at P<0.05

TABLE 5. Effect of body condition score on serum levels of some reproductive hormones, proteins, blood metabolites, blood antioxidants, and some trace elements in buffalo (Mean ±SEM).

Parameters				
T drameters	Low	Medium	High	
Progesterone (ng/ml)	$0.56{\pm}0.7^{b}$	$0.93{\pm}0.1^{a}$	$1{\pm}0.07^{a}$	
Estradiol (pg/ml)	$0.67{\pm}0.07^{b}$	$0.91{\pm}0.07^{a}$	$0.9{\pm}0.04^{a}$	
Total protein (g/dl)	5.1±0.3 ^b	6.4 ± 0.3^{a}	$6.4{\pm}0.2^{a}$	
Albumin (g/dl)	2.5±0.1 ^b	2.9±0.1 ^a	3±0.1 ^a	
Globulin (g/dl)	$2.7{\pm}0.2^{b}$	3.6 ± 0.2^{a}	3.5±0.1 ^a	
Albumin/globulin ratio	0.6 ± 0.05^{b}	$0.73{\pm}0.07^{ab}$	$0.83{\pm}0.06^{a}$	
Triglycerides (mg/dl)	62±2.6 ^b	74.2 ± 4.9^{a}	77.2±3.7 ^a	
Total cholesterol (mg/dl)	92.4 ± 5.4^{b}	112.9±5.5 ^a	119.7±5.6 ^a	
LDL (mg/dl)	124.3±5.4 ^a	57.1±4.4 ^b	55.1±2.8 ^b	
Glucose (mg/dl)	57.5±2.9 ^a	61.2±3.1 ^a	60.9±2.5 ^a	
SOD (u/ml)	53.6±3.9°	89.2±7.5 ^b	116.1±6.3 ^a	
MDA (nmol/ml)	12.3±0.5 ^a	8.8 ± 0.3^{b}	8±0.1 ^b	
Zn (umol/l)	0.11 ± 0.001^{b}	0.13 ± 0.003^{b}	$0.15{\pm}0.007^{a}$	
Cu(umol/l)	0.22 ± 0.01^{b}	0.3±0.01 ^a	$0.36{\pm}0.02^{a}$	

Superscripts with different letters within the same row differ significantly at P<0.05.

References

- 1. Arefaine, H. and Kashwa, M.A. Review on strategies for sustainable buffalo milk production in Egypt, *Journal of Biology, Agriculture and Healthcare*, **5**, 63– 67 (2015).
- 2. FAOSTAT. FAO Statistics Division; FAO: Rome, Italy; Available online: www.fao.org (2021).
- 3. United Nations, Department of Economic and Social Affairs, Population Dynamics. *World Population Prospects* (2019). Available online: https://population.un.org/wpp (2021).
- 4. Soliman, S.S., Abdoon, A.S., El-Toukhey, N.E., Kandil, O.M., Sabra, H.A. and Attia, M.Z. Transcript abundance of GAPDH, PGES, HSP 70, PPAR γ and SOD 2 mRNA genes expression during the different stages of reproduction in Egyptian buffaloes, *Journal of Egyptian. Veterinary Medicine Association*, **80** (2), 193 – 208 (2020).
- Soliman, S.S., Attia, M.Z. and Abdoon, A.S. Seasonal variation in ovarian functions in Egyptian buffalo and cattle, *International Journal of Pharm. Tech. Research*, 9 (6), 34-42 (2016).
- D'Occhioa, M.J., Ghuman, S.S., Neglia G., della Vallec, G., Baruselli, P.S., Zicarellic, L., Visintind, J.A., Sarkare, M., Campanilec, G. Exogenous and endogenous factors in seasonality of reproduction in buffalo: A review. *Theriogenology*, **150**, 186-192 (2020).
- Soni, N.A., Pandey, K., Kumar, A., Verma, A., Kumar, S., Gunwant, P., Phogat, J.B., Kumar, V. and Singh, V. Expression of MTNR1A, steroid (ERα, ERβ, and PR) receptor gene transcripts, and the concentration of melatonin and steroid hormones in the ovarian follicles of buffalo. *Domestic Animal Endocrinology*, **72**, 106371 (2020).
- Zandi, P. and Schnug, E. Reactive oxygen species, antioxidant responses and implications from a microbial modulation perspective. *Biology (Basel)*, 11(2), 155 (2022).
- Deepak, G., Ingole, S.D., Bharucha, S.V., Pharande, R.R., Dagli, N.R., Kharde, S.D. and Singodi M. Quantification of kiss peptin and progesterone concentrations in ovulatory and an ovulatory buffalo. *The Pharma Innovation Journal*, **11**(7), 3526-3530 (2022).
- Xulu, T.G., Ncobela C.N., and Kunene N.W. Influence of season and rangeland-type on serum biochemistry of indigenous Zulu sheep, *Open Agriculture*, 7(1) 455-464 (2022).
- 11. Laubenthal, L., Ruda, L., Sultana, N., Winkle, r J., Rehage, J., Meyer, U., Dänicke, S., Sauerwein, H. and Häussler, S. Effect of increasing body condition on oxidative stress and mitochondrial biogenesis in subcutaneous adipose tissue depot of non-lactating dairy cows. *Journal of Dairy Science*, **100**, 4976–4986 (2017).
- 12. Stefańska, B., Nowak, W., Oszmałek, E.P., Mikuła, R., Stanisławski, D., Kasprowicz-Potocka, M., Frankiewicz, A. and Maćkowia, P. The effect of body condition score on the biochemical blood indices and reproductive performance of dairy cows, *Annals of*

Animal Science, 16 (1), 129-143 (2016).

- Çolakoğlu, H.E., Yazlık M.O., Pekcan, M., Kaya U., Kaçar, C. and Vural, M.R. Impact of prepartum body condition score loss on metabolic status during the transition period and subsequent fertility in Brown Swiss dairy cows. *Journal of Veterinary Research*, 63, 375-382 (2019).
- Mberato, Y., Hamsun, M., Fatmawaty, S. and Mirajuddin. Effect of non-genetic factors on calving interval of swamp buffalo in Poso district, Indonesia. *Australian Journal of Basic and Applied Sciences*, 10(4), 187-192 (2016).
- 15. Faraz, A., Tauqir, N.A., Waheed, A., Ishaq, H.M., Hussain, S.M., Mirza, R.H., Bilal, R.M., Akbar M.A., and Nabeel, M.S. Haematological and biochemical profile of extensive kept male Barela (Camelus dromedarius) camel during breeding and non-breeding season. *Sarhad Journal of Agriculture*, **38** (4), 1184-1188 (2022).
- Noaman, V., Rasti, M., Ranjbari, A.R., Shirvani, E. Copper, zinc, and iron concentrations in blood serum and diet of dairy cattle on semi-industrial farms in central Iran. *Tropical Animal Health Production*, 44, 407–411 (2012).
- Abdoon, A.S., Soliman, S.S., Attia, M.Z., El-Toukhey, N.E., Kandil, O. M., Sabra, H.A. Seasonal variation in the number of ovarian follicles and hormonal levels in Egyptian Buffalo and cattle. *International Journal of Veterinary Science*, 9(1), 126-130 (2020).
- Worku, K., Kechero, Y. and Janssens, G.P. Measuring seasonal and agro-ecological effects on nutritional status in tropical ranging dairy cows. *Journal of Dairy Science*, **104**(4),4341–4349 (2021).
- Mengwei, L., Faiz-ul, H., Zhenhua, T., Yanxia, G., Xin, L., Lijuan, P., Huade, X. and Chengjian Y. Physiological, oxidative and metabolic responses of lactating water buffaloes to tropical climate of South China. *Veterinary Medicine and Science*. 7(5), 1696-1706 (2021).
- 20. Ahmad, M., Bhatti, JA., Abdullah, M., Javed, K., Din, R., Ali, M., Rashid, G., Ahmed, N., and Jehan, M. Effect of different ambient management interventions on milk production and physiological performance of lactating Nili- Ravi buffaloes during hot humid summer. *Livestock Research for Rural Development*, 29 (12), 1-18 (2017).
- Abel-Ghani, A.A., Solouma, A.M.A., Abd Elmoty, A.K.I., Kassab, A.Y. and Soliman, E.B. Productive performance and blood metabolites as affected by protected protein in sheep. *Journal of Animal Science*, 1(2), 24-32 (2011).
- 22. Costanzo, P.R., Suárez, S.M., Kozak, A.E., Knoblovits, P. Seasonal variations in sex steroids in a young male Population and their relationship with plasma levels of vitamin D. *The World Journal of Men's Health*, **40**(2), 308-315 (2022).
- 23. Xulu, T.G., Ncobela, C.N. and Kunene, N.W. Influence of season and rangeland-type on serum biochemistry of indigenous Zulu sheep. *Open Agriculture*, 7(1), 455–464 (2022).
- 24. Pandey, N., Kataria, N., Kataria, A.K. and Josh, i. A.

Ambient stress associated variations in metabolic responses of Marwari goat of arid tracts of India. *Journal of Stress Physiology and Biochemistry* **8**(3), 120-126 (2012).

- Miki, S., Balboula, A.Z., Yamanaka, K. and Takahashi, M. Effect of summer heat environment on body temperature, estrous cycles and blood antioxidant levels in Japanese Black cow. *Animal Science Journal*, 83, 394–402 (2012).
- 26. Abdoon, A.S.S., Al-Atrash, A.M.E., Soliman, SS., El-Sanea, A.M., Gamal el Din, A.A., Fahmy, HM. Lyophilized equine platelet-rich plasma (L-GF^{equina}) antagonizes the reproductive toxicity and oxidative stress induced by cyclophosphamide in female rats. *Journal of Ovarian Research*, **16**, 84(2023).
- Lallawmkimi, C.M. Impact of thermal stress and vitamin-E supplementation on heat shock protein 72 and antioxidant enzymes in Murrah buffaloes. Ph D Thesis. NDRI Deemed Univ. Karnal, India (2009).
- Teng, Y. and Liu R. Insights into potentially toxic effects of 4-aminoantipyrine on the antioxidant enzyme copper-zinc superoxide dismutase. *Journal of Hazardous Materials*, 262, 318–324 (2013).
- Preeti, L., Alhussien, M.N., Lakhanic, N., Jindala, R. and Nayyar, S. Seasonal variation in physiological responses, stress and metabolic-related hormones, and oxidative status of Murrah buffaloes. *Biological Rhythm Research*, **49**(6), 844–885 (2018).
- Chen, R., Lu, J., Guo, R., Mei, C., Guo, B., Li, W., Tsigkou, A., Shi, Z. Rectifying cow infertility under heat stress by immunization against inhibin and supplementation of progesterone. *Domestic Animal Endocrinology*, **80** (106726), 1-9 (2022).
- Das, G.K. and Khan, F.A. Summer Anoestrus in Buffalo – A Review. *Reproduction in Domestic Animals*, 45, 483–494 (2010).
- 32. Abd El-Razek, E.M.M. and Allam, T.S. Some biochemical parameters and hematological picture in cases of smooth inactive ovaries in buffalo-cows. *Alexandria Journal of Veterinary Sciences*, **60** (1), 83-92 (2019).
- Hafez, M.H. Serum Hormonal, Metabolic and minerals profile in normal cyclic and postpartum anestrus Egyptian buffaloes. *Alexandria Journal of Veterinary Sciences*, 60 (2), 102-108 (2019).
- 34. El-Sheikh, M., Mesalam, A.A., Kang, S.M., Joo, M.D., Soliman, S.S., Khalil, A.A.K., Ahn, M.J. and Kong, I.K. Modulation of apoptosis and autophagy by melatonin in Juglone-exposed bovine oocytes. *Animals.* 13(9), 1475 (2023).
- Mondal, M.K. and Samik, K.P. Haemato-biochemical profile in repeat breeding cross-bred cows. *Exploratory Animal and Medical Research*, 2(1), 60-65 (2012).
- 36. El-Khadrawy, H.H., Ahmed, W.M. and Hanafi M. Observations on repeat breeding in farm animals with emphasis on its control. *Journal of Reproduction and Infertility*, 2(1) 01-07 (2011).
- 37. Ahmad, I., Lodhi, L.A., Qureshi, Z.I. and Younis M. Studies on blood glucose, total proteins, urea, and cholesterol levels in cyclic and non-cyclic and

endometritic crossbred cows. *Pakistan Veterinary Journal*, **24**(2), 92-94 (2004).

- Damptey, J.K., Obese, F.Y., Aboagye, G.S., Ayim-Akonor, M. and Ayizanga, R.A. Blood metabolite concentrations and postpartum resumption of ovarian cyclicity in Sanga cows. *South African Journal of Animal Science*, 44(1)10-17 (2014).
- 39. Abdoon, A.S.S., Attia, M.Z., El-Toukhey, N.E., Kandil, O.M., Sabra, H.A. and Soliman, S.S. Effect of reproductive status and season on blood biochemical, hormonal and antioxidant changes in Egyptian buffaloes. *International Journal of Veterinary Science*, 9(1), 131-135 (2020).
- El-Wishy, A.B. The postpartum buffalo II. Acyclicity and anestrus. *Animal Reproduction. Science*, **97**, 216-236 (2007).
- 41. Prajapati, A.R., Dhami, A.J., Hadiya, K.K. and Patel, J.A. Influence of estrus synchronization protocols on plasma profile of progesterone, protein and cholesterol in acyclic Holstein Friesian crossbred cows. *Indian Journal of Veterinary Science & Biotechnology*, **13**, 5– 11 (2018).
- 42. Sirotkin, A.V., Makarevich, A.V., Makovicky, P. and Kubovicova, E. Ovarian, metabolic and endocrine indexes in dairy cows with different body condition scores. *Journal of Animal and Feed Sciences*, **22**, 316– 322 (2013).
- 43. Mouffok, C., Madani, T., Smara, L., Baitiche, M., Allouche, L. and Belkasmi, F. Relationship between body condition score, body weight, some nutritional metabolites changes in blood and reproduction in Algerian Montbeliad cows, *Veterinary World*, 4(10), 461-466 (2011).
- 44. Manzoor, A., Untoo, M., Zaffa, r B., Afzal, I., Fayaz, A., Dar, ZA. and Shafiq, S. Performance profile of dairy animals under compromise with dynamics in body condition score. A review. *Journal of Animal Health and Production*, 6 (3), 80 (2018).
- 45. Yaylak, E., Yenisey, C. and Seyrek, K. Effects of lameness, stage of lactation and body conditions score on some blood parameters in Holstein cows. *Asian Journal of Animal and Veterinary Advances*, 4(5), 245-251 (2009).
- 46. Aktaş, M., Ozkanlar, S., Uçar, O., Özkanlar, Y. E., Kaynar, O., Aytekin, I. Relationships between body condition score and some metabolic blood parameters in early lactating dairy cows. *Revue De Medecine Veterinaire*, **162**(12), 586-592 (2011).
- 47. Lake, S.L., Scholljegerdes, E.J., Hallford, D.M., Moss, G.E., Rule, D.C. and Hess, B.W. Effects of body condition score at parturition and postpartum supplemental fat on metabolite and hormone concentrations of beef cows and their suckling calves. *Journal of Animal Science*, 84(4), 1038–1047 (2006).
- 48. Derkx, M.M.S. The effect of body condition in the early dry period on the oxidative status of early lactation cows. *Utrecht University Student Theses Repository Home* (2021).

تأثير الحالة التناسليه وحالة الجسم والمكان على الهرمونات وبعض مكونات لايض في دم الجاموس المصري

سهام سمير سليمان، عمرو محمد محمد الصانع²، أميمه محمد توفيق قنديل³، أمل محمود أبو المعاطى⁴ وأحمد صبرى صلاح الدين عبدون⁵

قسم التكاثر في الحيوان والتلقيح الاصطناعي، معهد البحوث البيطريه، المركز القومي للبحوث

تم تصميم هذه الدراسة لمعرفة تأثير الحالة التناسليه، وحالة الجسم، والمكان على المستوى الهرموني، وحالة مضادات الأكسدة،والعناصر النادره،والتحليل البيوكيمائي والايض في الدم في الجاموس المصري. أجريت ثلاث تجارب: في التجربة 1: تم تقسيم الجاموس إلى مجموعتين حسب المحافظات (الفيوم أوالبحيرة). في التجربة 2: تم تقسيم الجاموسً إلى منتظم الشياع (ن = 29)، والحمل المبكر (ن = 43)، وخمول المبايض (ن = 12)، والتهاب بطانة الرحم (ن = 13). في التجربة الثالثة: تم تقسيم الجاموس إلى ثلاث مجموعات حسب درجة حالة الجسم (ضعيفه، متوسطة، وجيده). تم جمع عينات الدم من جميع الحيوانات لتحليل هرمون البروجسترون (P4) والاستروجين (E2) وفوق أكسيد ديسموتاز (SOD)، المالونديالدهيد (MDA)، البروتين الكلى (TP)، الألبومين (Alb)، الكوليسترول الكلى، الدهون الثلاثية، البروتين الدهني منخفض الكثافة، (LDL)، الجلوكوز، الزنك (Zn)، والنحاس (Cu). أشارت النتائج إلى أن مستويات P4 و E2 ومضادات الأكسدة و TP و Alb و Glob و A/G والجلوكوز والزنك والنحاس زادت بشكل معنوي (P<0.05) في الجاموس المربى في البحيرة (الدلنا) مقارنة بالفيوم (صعيد مصر). في الجاموس الحامل والمنتظم الشياع، زادت نسبة P4 وE2 ومضادات الأكسدة وTP وGlob وA/G والكوليسترول والدهون الثلاثية والجلوكوز وSOD والزنك والنحاس (P<0.05) مقارنة مع التي تعانى من خمول المبايض أو التهاب بطانة الرحم. علاوة على ذلك، اظهر الجاموس ذو الحاله الجسمانية الجيده والمتوسطة مستويات أعلى (P<0.05) من P4 وE2 وTP وdlb وgbb والكوليسترول الكلي والدهون الثلاثية والنحاس مقارنة بالمجموعه صاحبه درجة حالة الجسم الضعيفه. كانت مستويات MDA وLDL مرتفعة (P<0.05) في الجاموس ذو الحالة الجسمانيه الضعيفة. **الخلاصه**: كانت تركيزات مصل الدم من P4، E2، البروتين الكلي، الألبومين، الجلوبيولين، الكوليسترول الكلي، LDL، الدهون الثلاثية، الزنك، والنحاس هي العلامات البيوكيميائية الأكثر حساسية لتمثيل الحالة التناسليه للجاموس. كما أثرت درجة حالة الجسم والمكان على خصوبة جاموس.

الكلمات الدالة: الجاموس، المكان، الحالة التناسلية، درجة حالة الجسم، الهرمون، التحليل البيوكيمائي.