

CHANGES IN SOME HAEMATOLOGICAL AND BIOCHEMICAL PARAMETERS IN LOCAL BLACK GOATS DURING PREGNANCY

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ABSTRACT

The present study was aimed to assess and determine the effects of pregnancy on some biochemical and haematological parameters. This experiment was conducted using 16 pregnant of known gestational period and 4 non-pregnant does raised at commercial farm. Results revealed a non-significant differences between pregnant and non-pregnant does for all studied traits. With the advances of pregnancy, it was noticed an increase ($P<0.05$) in PCV, Hb, AST, ALT, Glucose and Cholesterol and a decrease in total protein and globulin. In conclusion, it can be indicated that physiological status impacts the traits under consideration.

Keywords: Protein; physiology; gestation period.

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التغيرات في بعض المعالم الدموية والكيموحيوية في الماعز المحلي الاسود خلال فترة الحمل

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المستخلص

لدراسة تأثير الحمل في بعض المعالم الدموية والكيموحيوية فقد تم استعمال 16 معزة حامل و4 معزات غير حامل مرياة في احد القطعان التجارية. تشير النتائج بانعدام الفروقات المعنوية بين النعاج الحامل وغير الحامل في جميع الصفات قيد الدراسة. كما لوحظ بان مرحلة الحمل قد صاحبه زيادة معنوية ($T > 0.05$) في حجم الخلايا المرصوصة، خضاب الدم، أسبارتيت ترانساميناز Ast، الانين ترانساميناز Alt، الكلوكوز والكوليستيرول وانخفاض في البروتين الكلي والكلوبيولين. وعليه يمكن الاستنتاج من هذه الدراسة بان المرحلة الفسيولوجية ذات تأثير معنوي على الصفات المدروسة.

الكلمات المفتاحية: حجم الخلايا المرصوصة، خضاب الدم، النعاج، المرحلة الفسيولوجية.

INTRODUCTION

Goat is one of the crucial ruminant animals to the human beings worldwide. In comparison to other ruminants, goats have significant characteristics such as their unique ability to adapt and survive under harsh environmental conditions. In Iraq, the indigenous goats with their population 1.3 million heads are raised mainly for production of meat and milk (19). However, in contrast to domestic sheep, limited researches have been attempted to categorize several performance and economic traits in the Iraqi indigenous goats (3). Physiological stages such as pregnancy and lactation have been indicated to impact metabolism in mammals (23,28). For instance, feed intake, body composition, energy and metabolism are changed in order to provide an acceptable source of nutrients for the development of the fetus (32). Furthermore, biochemical parameters of blood including free fatty acids, serum total protein, triglycerides and urea are substantial indicators of the nutritional and health conditions of the animals (22). Several previous studies about biochemical blood parameters have also shown alterations in activities of liver enzymes and metabolism during lactation and dry periods in ruminants (27, 41). Besides, studies of massive variations in the biochemical and haematological parameters between different goat breeds have been well characterized (7,43). The present study was aimed to investigate changes in some biochemical and haematological values for pregnant and non-pregnant local black goats during the gestation period.

MATERIALS AND METHODS

Location and experimental duration

The present experiment was carried out at the commercial farm located at Jom Jehany village. Summel region, Duhok governorate during the period from 1-8-2020 to 15-5-2021.

Animals and management: In the present study, 60 adult female goats (3-4 years old) with an average live body weight 43kg were used. The female goats were randomly distributed into four groups, each one with 15 does. The estrus for does were synchronized using sponge impregnated with progesterone (40 mg medroxyacetate (MAP))^A for 14 days. After sponge withdrawn, the first group as a

control was injected with normal saline (0.9%NaCl) (T1). The second group was injected intramuscularly with 250 IU hCG^B (T2). The third group was injected intramuscularly with 400 IU eCG^C (T3), and the fourth group was injected intramuscularly with 20µg GnRH^D (T4). At 35 days following mating, blood samples were taken for detection of pregnancy by measuring the progesterone level using Liaison Diasorin Kit^E. Then, 16 pregnant does and 4 non-pregnant does were chosen to determinate some changes in haematological and biochemical blood parameters.

A (40 mg medroxyacetate (MAP) SPAIN).

B hCG (Weser-HCG5000 IU,Germany).

C eCG (OVISER 5000 , SPAIN)

D GnRH (Gestar, GnRh OVER,Argentina).

E Progesterone Kit (Liaison DiaSorin kit, Germany).

Blood sampling and assay

To determine biochemical and haematological parameters, approximately 10ml of whole blood was collected from the jugular vein of each individual in the morning at 9AM before grazing at days 45, 90 and 35 post mating. For estimation of hemoglobin (Hb) concentration, packed cell volume (PCV), determination of N/L, 2.5ml out of 10ml of blood was collected in anticoagulant EDTA tubes. The remaining blood 7.5ml was transferred into non-anticoagulant sterile serum tubes. After 2hrs of leaving in the room temperature, the tubes were centrifuged for 15 minutes at 3000RPM. Then, the serum was pulled using micropipette and transferred into Eppendorf tubes. The serum tubes were then stored at -20°C until further analysis for glucose, total protein, globulin, albumin, Alt, Ast and cholesterol.

Determination of Hemoglobin

Concentration of hemoglobin (Hb) was assessed following Cyanmethemoglobin method (12) and was calculated according to the equation: Hemoglobin (g/dl) = (absorption of sample/ absorption of standard) *15g/d.

Quantification of Packed Cell Volume (PCV):

The PCV was measured using microhematocrit method. Firstly, heparinized capillary tubes were filled with blood samples and one end of the tubes was sealed with sticky material. Then, tubes were centrifuged at 10000 RPM for 5 minutes using specialized

PCV microcentrifuge. The percentages of PCV were calculated using micro-hematocrit capillary tube reader.

Determination of N/L: Blood smears were made by placing a small drop of sample onto clean slides. The smeared slides were then dried and stained with a hematologic Wright's stain (kitRef). Differential leukocyte counts were performed under a higher magnification (100X oil immersion objective) following a method described

Determination of blood biochemical Levels

Biochemical parameters were determined using kits from Roche Diagnostics Company (Germany), for analysis of total protein (TP: CAN 678), albumin (ALB: ACN413), globulin, Aspartate Aminotransferase (AST: CAN 687), Alanine Aminotransferase (ALT: ACN685), Glucose (GLU: ACN717) and Cholesterol (CHO:ACN798). Automated method using Biochemical auto analyzer Cobas C 501 in Ammer lab in Duhok.

Assessment of globulin

Quantity of globulin was measured by substrating albumin quantity from total protein.

Progesterone assay for detection pregnancy

To detect pregnancy, progesterone level was measured after 35 days of mating using Liaison Diasorin Kit (Germany).

Statistical analysis: Data collected from haematological and biochemical parameters were arranged in a (4X1) factorial layout including control treatment. Analysis of Variance in Completely Randomized Design was applied. Duncan's Multiple Range (F-Test) at ($p < 0.05$) within the SAS software was designated to find statistical significance amongst treatments (15, 37).

RESULTS AND DISCUSSION

Haematological parameters

In the present study, the overall means for the Packed Cell Volume (PCV) and hemoglobin (Hb) were $28.96 \pm 0.73\%$ and $9.60 \pm 0.23\text{g/dl}$ respectively (Table 1). Similarly, these values indicated in the present study are within the range (PCV: 22-38%) and (Hb: 8-12g/dl) as reported by previous studies for other breeds of goat (25, 33, 35, 30). Furthermore, as shown in Table 1, although the values of PCV and Hb numerically are higher in pregnant does than in non-pregnant ones, the statistical

differences among them were not significant. These findings are in accordance with observations recorded by others (29, 44), when they found no significant effects of pregnancy on Hb and PCV values in goats. The results show that goats have a remarkable ability to respond to physiological stresses during pregnancy. On the other hand, it seems from the Table 1, that both PCV and Hb values were significantly ($P < 0.05$) increased with advances of pregnancy. Such increase could be due to the great demand for the requirements of higher metabolic rate and for the oxygen during pregnancy. These rises may also be related to increased volume of red blood cells (RBC) throughout pregnancy. These findings are compatible with those described for PCV by previous studies (18,4). Nevertheless, other researches in ewes (2, 10) contrarily observed that PCV values were decreased with advancing pregnancy. Regarding Hb values, similar results were observed by other researchers (40, 17, 11, 9, 1) when they recorded higher values of Hb in ewes at the end of pregnancy, With respect to neutrophil: lymphocyte ratios (N/L %), results indicated as shown in Table 1 that N/L% was increased numerically in the late pregnancy. Similarly, highest means of neutrophils were found in the Rembi ewes during late pregnancy (2). These results are similar to those stated by others (1, 9), when they noticed that neutrophils were highest in pregnant ewes in comparison to the other values of white blood cells (WBC). This increase in neutrophils may be due to the increase in hormonal secretion that occurs under stressful conditions, and the negative energy balance that occurs in ewes during late pregnancy. Stress can lead to leukocytosis due to the increase and release of bone marrow neutrophil reserves, induced by glucocorticoids (42). Moreover, others (14) indicated that the stress conditions possibly stimulate the release of some factors such as Colony Stimulating Factors (CSF) and Leukocytosis inducing Factor (LIF), which are known to increase metabolism of blood cells and increase haemopoietic activities. Additionally, other studies (21) pointed out that the negative energy balance activates the function of the adrenal glands, which can lead

to an increase in the levels of catecholamines, cortisol and endorphins.

Table 1. Changes (Mean \pm SE) in some haematological parameters in black goats during pregnancy

Effect	N	PCV %	Hb g/dl	N	L	N/L %
overall mean	60	28.96 \pm 0.73	9.60 \pm 0.23	22.40 \pm 0.69	77.81 \pm 0.69	0.29 \pm 0.01
Non-pregnant	12	27.91 \pm 1.51 a	9.24 \pm 0.49 a	24.41 \pm 1.37 a	75.66 \pm 1.36 a	0.32 \pm 0.02 a
pregnant	48	29.22 \pm 0.83 a	9.69 \pm 0.27a	21.89 \pm 0.78 a	78.35 \pm 0.78 a	0.28 \pm 0.01 a
Period /day						
45 (early pregnancy)	20	24.50 \pm 0.91 b	8.15 \pm 0.29 b	21.75 \pm 1.05 a	78.30 \pm 1.04 a	0.28 \pm 0.01 a
90 (mid pregnancy)	20	30.05 \pm 1.03 a	9.98 \pm 0.33 a	21.90 \pm 1.29 a	78.70 \pm 1.26 a	0.28 \pm 0.02 a
135 (late pregnancy)	20	32.35 \pm 1.18 a	10.67 \pm 0.38 a	23.55 \pm 1.26 a	76.45 \pm 1.26 a	0.31 \pm 0.02 a

Means with different letters refer to the statistical significance at ($p < 0.05$). While, means with the same letters indicate that they do not differ significantly

Blood biochemical profile

The blood biochemical profile measured during the experiment is shown in Table 2. The overall mean of total protein, albumin and globulin concentration averaged 6.93 \pm 0.10, 2.86 \pm 0.04 and 4.08 \pm 0.10 g/dl, respectively. The values of protein concentration observed in the present study are compatible with those recorded for goats by other investigations (25). Also, it appears from Table 2, that physiological conditions of does (pregnant vs non-pregnant) have not shown statistical significance ($P > 0.05$) in respect of means of total protein and their fractions. Compatibly, similar results concerning the values of total protein and globulin were found between pregnant and non-pregnant of Qatari goats (44, 45). Besides, it seems from Table 2, that the impact of period on total protein and globulin was significantly ($P < 0.05$) lower in mid (6.75 \pm 0.12, 3.78 \pm 0.12g/dl) and late pregnancy (6.57 \pm 0.16, 3.79 \pm 0.16) as compared to early pregnancy (7.47 \pm 0.19, 4.65 \pm 0.17g/dl). Such decrease in the concentrations of maternal serum protein could be due to an increased growth of the fetal muscles, particularly the utilization of amino acids from maternal circulation (5). Similar results were found by Piccione et al. (34). In contrast to our results, Zebari et al. (46) and Juma et al. (24) found that total proteins in serum were significantly ($P \leq 0.05$) higher in late pregnancy as compared with early pregnancy. Blood serum albumin did not differ significantly among the stages of pregnancy, which agree with the results observed by Zebari et al. (46). However, Kumar et al (29) recorded no significant effects of pregnancy stages on

different metabolites in terms of blood haemato-biochemical profile. The overall means of glucose and cholesterol were 48.05 \pm 1.20 and 67.48 \pm 2.36 mg/dl respectively (Table 2). The values obtained herein are within the normal range recorded for goats (27,36). It has been indicated that the physiological conditions of does have no significant impact on glucose as well as cholesterol, although its well known that fetal requirements for energy are supplied primarily by maternal glucose (13). On the contrary, incompatible information were observed by Abdul-Rahaman et al (45) when they found that glucose and cholesterol were significantly increased ($P \leq 0.05$) in pregnant and decreased in non-pregnant goats. Additionally, significant effect ($P < 0.05$) of period on glucose and cholesterol was observed (Table 2). High values of glucose and cholesterol were found during mid (54.95 \pm 1.13, 82.20 \pm 4.18g/dl) and late pregnancy (50.40 \pm 1.82, 65.95 \pm 3.04g/dl) as compared with early pregnancy (38.80 \pm 1.29, 54.30 \pm 2.10 g/dl). In goats, it has been reported that lipids profile in blood serum during late pregnancy characterized by an increase in concentrations of triglycerides and total cholesterol (39). This could be due to the reduced responsiveness of target tissues to insulin, while an increased mobilization of lipid biomolecules from adipose tissue, which could provide new sources for growth of fetus (23), . Similar results were observed in other studies (5, 34) when they found significant increases in total cholesterol of blood serum at pregnancy and dry periods in goats. It's well known that cholesterol is vital to protect fluidity and

integrity of membrane structure and it also plays significant role in metabolism as a precursor of some vitamins, bile acids and steroid hormones (20). In ewes during late pregnancy, insulin responsiveness is decreased which lead to decrease in glucose and uptake by muscle and fat tissues. Besides, the outcome of hormones such as glucocorticoids, adrenaline and adrenocorticotrophic during the pregnancy is increased for breakdown glycogen in the liver. These released hormones perform considerable roles in the metabolism of amino acids from proteins and the associated conversion of α -ketoacids to glucose (8,39). Ast and Alt averaged 91.85 ± 4.63 IU/L and 18.11 ± 0.77 IU/L, respectively, and the differences among

pregnant and non-pregnant were not significant ($P > 0.05$) (Table 2). It indicates from the results, that the effect of period on AST and ALT was significantly ($P < 0.05$) higher during late pregnancy (127.80 ± 8.67 , 20.45 ± 1.41 IU/L) as compared with mid pregnancy (74.30 ± 3.07 , 14.85 ± 0.85 IU/L) respectively (Table 2). Similar results were observed by Antunovic et al. (6); El-Ghoul et al. (16); Juma et al (24); Stojevic et al. (41) and Zebari et al. (46). An increase in the activities of enzymes (AST and ALT) could be due to the basic requirement for amino acids in milk production in late pregnancy or may be due to the increases in metabolism in the liver during the specified stages (26, 31).

Table 2. Changes (Mean \pm SE) in some biochemical parameters in goats during pregnancy

Effect	N o	AST IU/L	ALT IU/L	Total protein g/dl	albumin g/dl	Globulin g/dl	Glucose mg/dl	Cholesterol mg/dl
overall mean	60	91.85 ± 4.63	18.11 ± 0.77	6.93 ± 0.10	2.86 ± 0.04	4.08 ± 0.10	48.05 ± 1.20	67.48 ± 2.36
Non-pregnant	12	93.75 ± 7.48 a	18.16 ± 4.46 a	7.13 ± 0.16 a	2.77 ± 0.08 a	4.40 ± 0.14 a	47.58 ± 2.58 a	70.58 ± 4.17 a
pregnant	48	91.37 ± 5.51 a	18.10 ± 0.92 a	6.88 ± 0.12 a	2.88 ± 0.05 a	3.99 ± 0.12 a	48.16 ± 1.37 a	66.70 ± 2.76 a
Period /day								
45 (early pregnancy)	20	73.45 ± 3.65 b	19.05 ± 1.42 a	7.47 ± 0.19 a	2.84 ± 0.06 a	4.65 ± 0.17 a	38.80 ± 1.29 c	54.30 ± 2.10 c
90 (mid pregnancy)	20	74.30 ± 3.07 b	14.85 ± 0.85 b	6.75 ± 0.12 b	2.96 ± 0.06 a	3.78 ± 0.12 b	54.95 ± 1.13 a	82.20 ± 4.18 a
135 (late pregnancy)	20	127.80 ± 8.67 a	20.45 ± 1.41 a	6.57 ± 0.16 b	2.77 ± 0.09 a	3.79 ± 0.16 b	50.40 ± 1.82 b	65.95 ± 3.04 b

Means with different letters refer to the statistical significance at ($p < 0.05$). While, means with the same letters indicate that they do not differ significantly

Conclusion

It could be concluded from the current study that haematological and biochemical imbalances are unlikely to happen in pregnant and non-pregnant local goats. On the other hand, Late and mid pregnancy period is associated with increases in PCV, HB, AST, ALT, glucose, cholesterol and decreases in total protein and Globulin.

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