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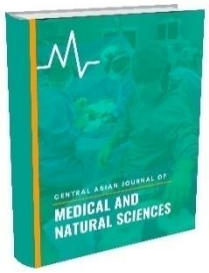

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Reproduction Statuses Impact Some Biochemical Parameters in Female Awassi Sheep in Al Diwaniyah Province / Iraq

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Abstract: Reproduction status consider a great modifier of metabolism status of animal. Measuring biochemical blood parameters is an important tool to evaluate animals health. This project aimed to evaluate the impact of pregnancy and lactation statues on some variations of blood including TC, TG, lipoproteins (HDL, LDL and VLDL), total protein and some of hepatic enzymes including ALT, AST and ALP.

Approximately 5-10 ml of venous blood were collected from clinically healthy awassi ewes. Enzymatic colorimetric procedure was used to estimate TC, TG and lipoproteins values and VLDL value was calculated. Hepatic enzymes activities were estimated by utilizing reflation apparatus. Significantly higher TC, TG, HDL, LDL , VLDL in serum were observed for pregnant group compared with control ewes. lactate group presented a significant decreased in TC at $p \leq 0.01$, TG, LDL, VLDL at $p \leq 0.05$ compared with their concentration in pregnant group. a significant change was presented of AST level and ALP level at $p \leq 0.01$ and $p \leq 0.0001$ in pregnant group comparing with control ewes. ALT didn't show significant differ between control and lactate groups. Under similar conditions of management and environmental nutrition , different reproduction (pregnant and lactate) statuses play a significant role in alteration lipoproteins and hepatic enzymes levels in Awassi ewes.

Key words: Awassi ewes, reproduction status, hepatic enzymes.

Introduction

Awassi sheep have height ability to afford poor nutrition's and cruel environmental, they consider as numerous number domestic kind in Iraq. These sheep's breed for multi purposes including wool, meat and milk production (Al-Hadithy, 2013; Talafha and Ababneh, 2011). Animal health status is based on assessing biochemical parameters of blood serum, animals with good health better to bred for future

offspring (Ramesh *et al*., 2019). Biochemical values of blood are an important set of tool to diagnose the health of animals (Van Saun, 2000). Which shows variations according to physiological factors that affect a variable levels of biochemical values (Roubies *et al.*, 2006). One of valuable blood parameters is lipids because they have a critical role in the body function, they serve as metabolic fuels, energy source and hormone precursors and documented a structural compounds of cell membrane (Musa, 2020). Lipoproteins transport lipids in serum composed protein complex. Physiological functions and origin places of lipoproteins were determined according to the shared protein in ruminants, composition and secretion of lipoproteins which they are consider as a main factors to control the metabolism of lipids in organs (Bauchart, 1993). Significant variation in lipids concentration lead to cause systemic disorders in influenced animals. Concentrations of triglycerides, carbohydrates, cholesterol and lipoproteins including very low density lipoprotein VLDL and height density lipoprotein HDL were decreased in post-partum period compared with late pregnancy (Fair *et al*., 2014). Reproduction statuses (pregnancy and lactate) have demonstrated to cause significant alterations in biochemical values and modifying metabolism in all animals (Iriadam, 2007; Karapehlivan *et al*., 2007). Maternal tissue is implicated in supplying energy during pregnancy in the production processing that could impact blood serum variables (Yokus *et al.*, 2006). It has been determined that lipoproteins, triglycerides and total cholesterol were increased in late pregnancy phase (Schlumbohm *et al*., 1997) because increasing fatty acid mobilization from adipose tissue and making new provenance for fetal growth (Iriadam, 2007). Relative to the metabolism of protein during the end of gestation in sheep was observed the using amino acid in the fetal muscles to synthesis the protein (Antunovic *et al.*, 2011) about 80% of blood metabolites were utilized to synthesis the milk in mammary gland of secretary cells during lactation stage (Karapehlivan *et al*., 2007). In addition to determined levels of lipids and lipoproteins during reproduction phases, significant alteration of liver enzymes levels were occurred. Aspartate transaminase (AST) enzyme was determined in skeletal muscle, cardiac, red blood cells and hepatocytes while alanine transaminase (ALT) is only identified in hepatocytes. alkaline phosphatase (ALP) enzyme has been expressed in placenta, bone and biliary ducts of lining cells (Copeland, 2023; Evans, 2009). These enzymes are utilized to assess hepatic functions. Even though some researchers develop their studies in enzymology field of sheep in Iraq, it is still a few information were identified of liver enzymes in awassi sheep in Iraq specially in Al Diwaniyah province. This work was conducted to establish baseline values for several biochemical of blood serum variables under different reproduction statuses in awassi sheep in Al Diwaniyah province of Iraq.

Materials and methods

Animals

Clinically healthy (thirty six) of female awassi sheep aged 2-3 years were chosen to assess reproduction statuses impact on some lipids lipoproteins and enzymes levels. These sheep's were divided into three groups including

Control group: 12 Awassi ewes non pregnant were served as a control group.

Pregnant group: 12 Awassi ewes were pregnant stage

Lactation group: 12 Awassi ewes were during lactation stage.

Blood samples collection

Approximately 5-10 ml of venous blood specimens were drawn from each female sheep placed in plain tubes to obtain serum samples to estimate biochemical parameters for this aim, samples were standing in slop position at room temperature following centrifugation at 3000 rpm for 5 minutes.

Biochemical variables measurements

Obtained serum samples were directly used for analysis cholesterol level was calculated using enzymatic colorimetric methods via commercial kit (Bio IABS, France). Triglycerides (TG) level was estimated following the methods of (Thomas *et al*., 2000). Commercial kits (Bio IABS, France) were used to determine the levels of HDL and LDL of serum blood samples using enzymatic colorimetric method .VLDL concentration was measured by dividing TG level on 5.

Remaining of serum was used to estimate AST, ALT and ALP enzymes levels following colorimetric methods as described by (Lee *et al*., 2003) via applying Reflotron analyzer apparatus

Statistical analysis

Obtained data of current study were statistically performed throug using ANOVA (one way) using software (Graph pad prism (version 8). All results were showed as means ± S.E. Differences between compared group were presented as significant value (p- value equal 0.05 or less than this level).

Results

Total cholesterol level was significantly increased up to (80.90±3.81) in pregnant ewes compared with control and lactate groups (55.60±1.5 and 52.50±1.84, respectively) as shown in Table (1) and Figure (1A). Triglycerides level does not show significant differ between lactate group (96.16±8.33) and control group (89.83±4.07) while it shows a significant differ between pregnant group and lactate group at p≤0.05 (Figure 1B). HDL level shows significant changes (25.16±0.81) in pregnant group compared with control group (21.03±0.98) while doesn't show significant changes with lactate group (17.73±0.85) as shown Table (1), Figure (1C). Lactate group doesn't show significant changes in LDL and VLDL levels (Table 1) while pregnant group shows significant changes of LDL level at (p≤0.05) and VLDL was change at (p≤0.0001) compared with control group (Figure 1D and E)

Groups	Total Cholesterol (mg/dL)	Triglycerides (mg/dL)	HDL (mg/dL)	LDL (mg/dL)	VLDL (mg/dL)
Control	55.60±1.51 a	89.83±4.07 a	21.03±0.98 a	18.00±3.25 a	17.96±0.81 a
Pregnant	80.90±3.81 b	130.17±3.72 b	25.16±0.81 b	32.72±6.62 b	24.60±1.91 b
Lactate	52.50±1.84 b	96.16±8.33 a	17.73±0.85 ab	15.53±1.82 a	19.23±1.66 a

Table 1: The Level Of Biochemical Variables In Awassi Ewes With Different Physiological Conditions.

In The Same Column, Different Letters Statistically Represents A Significant Differ (P≤0.05) While Similar Letters Referred To Non-Significant Values (P≥0.05).

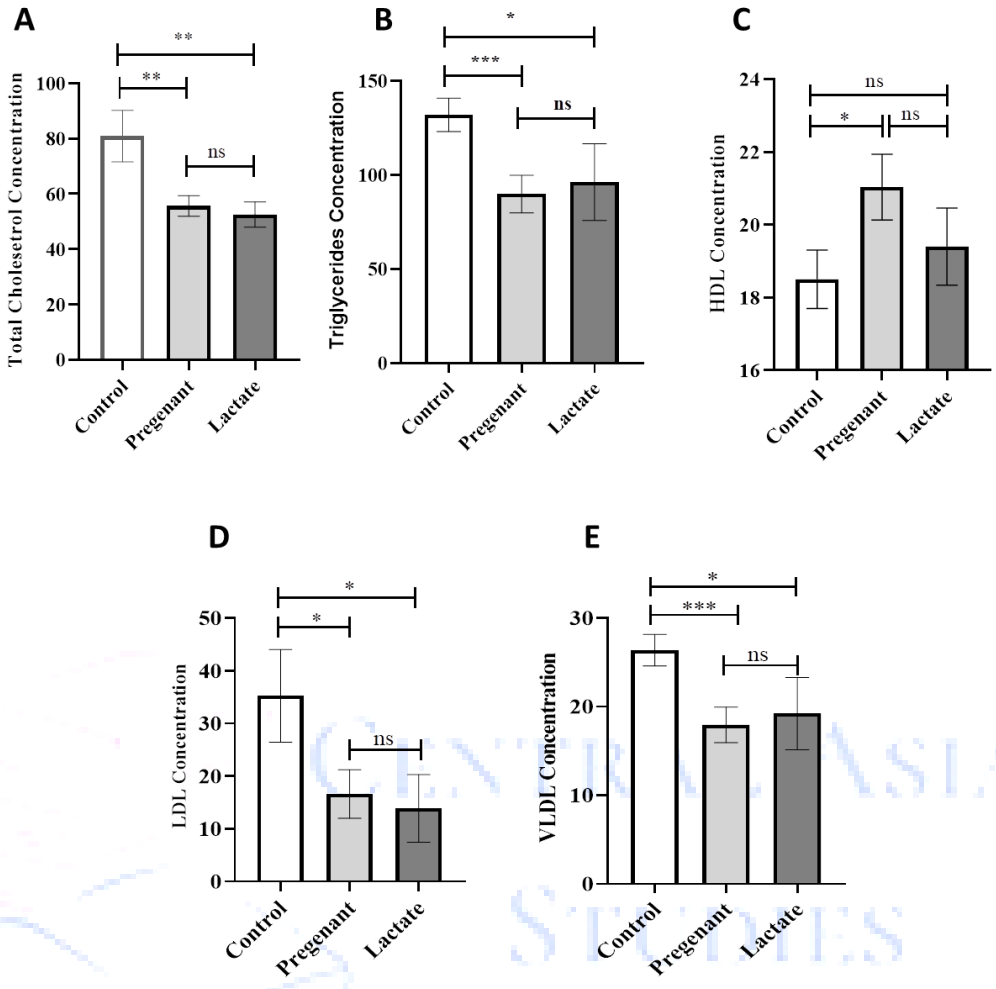

Figure 1: Level Of Biochemical Parameters In Awassi Ewes With Different Physiological Conditions.

*(A) Tc, (B) Tg, (C)Hdl, (D) Ldl, (E) Vldl. Statically Markers Presented As *(P≤0.05), **(P≤0.01),***(P≤0.001) And Ns(P≥0.05).*

Total protein level was down-regulated up to (6.33±0.3) in pregnant. While, a significant increasing was observed in total protein in lactate group (7.83±0.04) compared with its level in control group (7.50±0.04) at (p≤0.05) as shown in Table (2) and Figure (2A). The values of measured enzymes of liver were described in Table (2). ALT activity was decreased in pregnant group (23.00±1.57) while there is no significant alter was obtained between control group and lactate group Figure (2B). AST activity was (148.8±3.39) with significant change at (p≤0.01) in pregnant stage compared with control group (121.5±3.36) Figure (2C). However, there is no significant increase in lactate group (127.71±3.36) compared with control group. There is no significant differ between pregnant group and lactate group in ALP activity was significantly increased in pregnant stage (334.3±7.36) compared with control group (122.7±3.13) at (p≤0.0001) as well as significant change was observed in lactate group (257.7±2.95) at (p≤0.0001) compared with their control Table (2) and Figure (2D).

Groups	Total Protein (mg/dL)	ALT (U/L)	AST (U/L)	ALP (U/L)
Control	7.50±0.04 a	32.83±0.94 a	121.5±3.36 a	122.7±3.13 a
Pregnant	6.33±0.03 b	23.00±1.57 b	148.8±3.39 b	334.3±7.36 b
Lactate	7.83±0.04 a	20.17±1.30 a	127.7±2.10 a	257.7±2.95 b

Table 2: Activity Level Of Alt, Ast And Alp Enzymes Of Various Reproduction Statuses In Awassi Ewes.

In The Same Column, Different Letters Statistically Represents A Significant Differ (P≤0.05) While Similar Letters Referred To Non-Significant Values (P≥0.05).

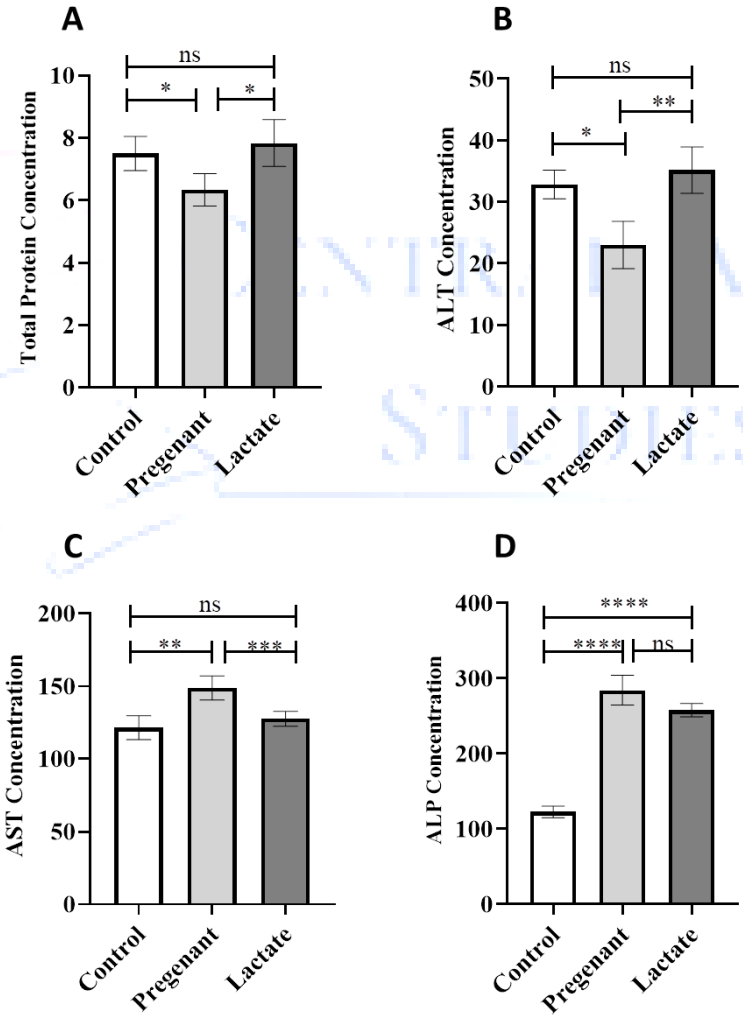

Figure 2: Activity Level Of Alt, Ast And Alp Enzymes Of Various Reproduction Statuses In Awassi Ewes.

*(A) Total Protein, (B) Alt, (C), Ast And (D) Alp. Statically Markers Presented As *(P≤0.05), **(P≤0.01), ***(P≤0.001), ****(P≤0.0001) And Ns(P≥0.05).*

Discussion

The significant high level of total cholesterol and triglyceride were determined during pregnancy stage compared with non-pregnant group lactate group. this significant increase support the results of (Hayyawi *et al.*, 2021) who determine increasing level of TC and TG in early and end stage of pregnancy. TG concentration is under complex factors controls observed decrease during lactate stage could be related to increases up taken of cholesterol` from tissues that involved in the synthesis of the milk as a consequences of responsiveness of insulin compared to the end stage of pregnancy (Nazifi *et al*., 2002) as well as TG plays an essential role in milk synthesis (Nazifi et al., 2002). Finding of current assay similar to (Antunovic *et al*., 2011) finding in terms of TC and TG when they find the level of TC and TG significantly down-regulated in lactate group. It has been observed that TC and TG were highly decreased in lactating group compared with their control while no significant observation was noticed in HDL and LDL levels in both groups (lactating and non-lactating) ewes awassi (Musa, 2020). In contrast in present study, HDL, LDL and VLDL were significantly increased in pregnant group and non-significant increase in lactate ewews compared with control ewes according to HDL and VLDL concentrations. VLDL lipoproteins mainly transports endogenous triglyceride that one synthesized in the liver . the ability of cattle liver to secret VLDL is very low paralleled to its ability to absorb fatty acid and esters (Mazur et al., 2009). It was noted that the low concentration of fats and lipoproteins may be related to the formation of milk and may be due to the difference in the ability to feed and metabolism between pregnancy and non-pregnant awassi ewes. As a result under the same environmental conditions, the physiological state of the body and the management system. Lactation phase play a major role in reducing the concentration of these fats. Several studies conducted on this topic revealed that low levels of nutrition may reduce lipoprotein concentrations (Fujita *et al*., 1996). Present study consistent with findings of (Ramesh *et al*., 2019; Sharma *et al*., 2015; Talawar *et al*., 2016) studies when they find the level of total protein is significantly decrease in pregnant group compared with lactating group. this decrease due to increasing the growth of fetal and utilizing the amino acids from circulating protein in maternal blood to synthesis the muscles of fetal (Antunovic *et al*., 2011). Significant evaluate of protein level in lactation stages could be related to lower level of globulin (El-Sherif and Assad, 2001). In addition, lactating ewes need high energy to synthesis the milk during early phase of lactation as confirmed by (Bremmer *et al*., 2000). In contrast, total protein concentration was up-regulated higher in pregnant one compared with its level in lactating ewes (Karapehlivan *et al*., 2007) while another study did not find any impact of physiological status on total protein concentration (Sarmin *et al*., 2021).

The effect of reproduction statuses in ALT.AST and ALP concentrations were somewhat controversial. ALT and AST concentrations were increased in pregnancy stage (Jovanovic *et al*., 1997). In present study there is significant changes were observed in hepatic enzymes. Current results observed decrease in ALT level while an increased in AST concentration in pregnant stage where ALP was significantly increased in pregnant group but there is no significant different was obtained between pregnant and lactate groups. While the study of Yokus and Cakir (2006) observed high activity of ALP level in mild and late stages of pregnancy that ALP activity in lactation. Yaralıoğlu Gürgöze *et al*. (2009) observed ALP level increased in lactation stage as well as current results find increasing in ALP level in lactation status compared with control group $p \leq 0.0001$. This increase could related to increase bone lsoenzymes production (Yaralıoğlu Gürgöze *et al.*, 2009) . Al-Hadithy (2013) noticed significant differ in enzymes levels in all studies group according to reproduction status. However, the serum enzymes levels did not find any significant differ between studied group (Ramesh *et al*., 2019) increase hepatic metabolism leads to increase AST and ALT activity (Antunovic *et al.*, 2011). However, the differences between observation of current study and the findings of other studies could be related to genetic factors or individuals variation. They could be related to the feeding system

furthermore the impact of heat stress, nutritional treatment and food type can be altered the concentration of liver enzyme to up level (Abdel-Samee *et al*., 2023).

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