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## Activity of transaminase enzyme and testosterone hormone in blood of Awassi rams during different season

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## ABSTRACT

**Objective:** To monitor the levels of aspartate transaminase (AST) and alanine transaminase (ALT) enzymes and testosterone hormone in months and seasons of Iraq. **Methods:** In this experiment, 20 (2.0–3.5 years old) Iraqi Awassi rams were used which were housed in semi opened shade. Blood samples (2–3 mL) were collected once a week for over period 12 mo. AST and ALT activities were measured by using colorimetric method, and testosterone analysis was performed with an automatic analyzer. **Results:** The activities of both AST and ALT enzymes were increased significantly ( $P < 0.05$ ) during summer, on monthly basis, June showed the significantly highest value ( $P < 0.05$ ). The present study revealed that testosterone in autumn and summer was recorded significantly highest ( $P < 0.05$ ) in comparison with other seasons and on monthly basis the significant ( $P < 0.05$ ) highest level of testosterone was found between September and August months. **Conclusions:** AST and ALT enzymes are the highest during summer, and on monthly basis they are the highest in June, while testosterone level is recorded highest in autumn season, and on monthly basis November shows the highest value.

## 1. Introduction

The secretion of testosterone is activated by external stimuli like smell and behavior of sheep and androgen secretion is directly correlated with the production of pheromones[1]. Enzymes in cells are needed to their functions. Any defect that occurs in the cell leads to the exit of enzymes abroad and measuring their activity has a role in the knowledge of cell safety and has a role in determining the extent of infection of the tissue disease[2]. The present enzymatic activity has a role in the diagnosis of animal diseases[3]. The changes that occur in normal rates of enzymes get as a result of a change in cell permeability or poor blood circulation or cell death[4]. Chronic liver disorders and the use of excessive stimulants during the normal growth of bones of young animals all lead to an increased level of alkaline phosphatase (ALP) enzyme[2]. This is added by Coppo's[5] discussion that physiological changes due to the progress of age, sex, reproductive status, nature of food and exercise have a role in the

occurrence of a change in this enzyme. Thermal changes, especially temperature rise, have not only a role in influencing the activity of enzymes and thus the cell's performance in physiological[6,7] and immunological terms[8,9]. In addition, seminal plasma enzymes are considered to be of significant importance to quality of semen[10]. Routine blood and semen tests are not alone enough to assess sperm susceptibility, especially when semen is used for artificial insemination purposes[11]. During the breeding season there was an improvement in the reproductive characteristics and biochemical analyzes of the blood serum, where a relationship was found between the enzymatic activity of both alanine transaminase (ALT) and ALP and the reproductive function[12]. This is confirmed by many studies[13–16]. There was also a significant relationship between total fat, cholesterol and enzymatic properties of the

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serum[12]. Analysis of enzymatic activity has a role in the extent of susceptibility of male cell to fertilization because it measures the effectiveness of the cell membrane sperm[13–17]. It was noted that increased sperm abnormalities led to a significant increase in the concentration of enzymes, but on the other hand decreased when the concentration of sperm and that these enzymes vary according to the types of rams[13]. More and more researches have shown that testosterone is essential for the expression of male sexual activity, but also for the development and maintenance of sexual behavior in rams[18]. The secretion of this hormone is activated by external stimuli like smell and behavior of sheep, and androgen secretion is directly correlated with the production of pheromones[1] and Deniz *et al*[19] found negative relationships were found between sperm concentration and aspartate transaminase (AST) and ALT ratio.

## 2. Materials and methods

In this experiment, 20 (2.0–3.5 years old) Iraqi Awassi rams were used which were housed in semi opened shade, provided with concentrated diet given at the rate of 0.75 kg in two-day occasions supplemented with straw and green food, and were kept under identical conditions of management, feeding and watering throughout the study period which started from December 2015 until November 2016. Rams were subjected to careful clinical examination to ensure that they were in good health without suffering from any disease. Blood samples (2–3 mL) were collected once a week for a whole year. The collected blood was drawn from the jugular vein and was centrifuged at 3 000 r/min for 10 min, then the sera were kept frozen ( $-20^{\circ}\text{C}$ ) until they were utilized for enzymes and hormone. AST and ALT activities were measured by using colorimetric method according to Reitman *et al* and Belfield *et al*[20,21]. Testosterone analysis was performed with an automatic analyzer Tossoh System (Hitachi Boehrering Mannheim, 912 Automatic Analyzer) using specific kits.

Data on the effect of months and seasons on the enzymes and testosterone hormones were analyzed using two-way ANOVA with the general linear model method of the SAS[22]. Comparisons between various percentage means were analyzed using Duncan multiple range test following significant *F*-test in ANOVA[23]. All statistical procedures were performed at 95% confidence level, and it meant significant probability ( $P<0.05$ ).

**Table 1**

Seasonality and monthly changes in AST, ALT, testosterone in blood of Awassi rams (means $\pm$ SE).

Item	Winter			Spring			Summer			Autumn		
	December	January	February	March	April	May	June	July	August	September	October	November
AST enzyme (iU/mL)	29.31 $\pm$ 0.23 <sup>b</sup>	20.13 $\pm$ 0.21 <sup>c</sup>	21.34 $\pm$ 0.23 <sup>c</sup>	24.40 $\pm$ 0.33 <sup>c</sup>	18.26 $\pm$ 0.42 <sup>d</sup>	22.09 $\pm$ 0.31 <sup>c</sup>	36.69 $\pm$ 0.25 <sup>a</sup>	33.23 $\pm$ 0.33 <sup>a</sup>	34.25 $\pm$ 0.27 <sup>a</sup>	28.21 $\pm$ 0.26 <sup>b</sup>	22.09 $\pm$ 0.19 <sup>f</sup>	20.24 $\pm$ 0.20 <sup>f</sup>
ALT enzyme (iU/mL)	9.24 $\pm$ 0.19 <sup>e</sup>	8.24 $\pm$ 0.17 <sup>c</sup>	8.17 $\pm$ 0.15 <sup>c</sup>	8.28 $\pm$ 0.14 <sup>c</sup>	7.21 $\pm$ 0.12 <sup>c</sup>	6.21 $\pm$ 0.11 <sup>c</sup>	18.67 $\pm$ 0.07 <sup>a</sup>	16.23 $\pm$ 0.10 <sup>a</sup>	17.32 $\pm$ 0.02 <sup>a</sup>	12.11 $\pm$ 0.16 <sup>b</sup>	11.87 $\pm$ 0.15 <sup>b</sup>	13.25 $\pm$ 0.02 <sup>b</sup>
Testosterone (ng/mL)	2.11 $\pm$ 0.03 <sup>d</sup>	2.09 $\pm$ 0.10 <sup>d</sup>	2.02 $\pm$ 0.07 <sup>d</sup>	3.21 $\pm$ 0.09 <sup>c</sup>	3.90 $\pm$ 0.11 <sup>c</sup>	2.23 $\pm$ 0.08 <sup>cd</sup>	5.71 $\pm$ 0.09 <sup>b</sup>	5.61 $\pm$ 0.11 <sup>b</sup>	5.99 $\pm$ 0.10 <sup>b</sup>	9.69 $\pm$ 0.12 <sup>a</sup>	8.03 $\pm$ 0.09 <sup>a</sup>	8.23 $\pm$ 0.08 <sup>a</sup>

Mean values in the same rows with different superscripts differ significantly ( $P<0.05$ ).

## 3. Results

The study results showed significant effect ( $P<0.05$ ) of different season on enzymatic activities (Table 1), and the activities of AST [(33.64 $\pm$ 0.23) iU/mL] and ALT [(16.42 $\pm$ 0.33) iU/mL] in summer were higher significantly ( $P<0.05$ ) than other seasons: winter [AST (26.41 $\pm$ 0.25) iU/mL, ALT (9.10 $\pm$ 0.26) iU/mL], autumn [AST (23.17 $\pm$ 0.24) iU/mL, ALT (12.68 $\pm$ 0.22) iU/mL] and spring [AST (21.56 $\pm$ 0.565) iU/mL, ALT (7.03 $\pm$ 0.19) iU/mL]. In addition, the activity of AST in spring was higher significantly ( $P<0.05$ ) than winter, and the activity of ALT in autumn was more significant ( $P<0.05$ ) than winter and spring season. On the other hand, the activity of AST in autumn did not differ significantly in comparison with spring and winter, and the activity of ALT in winter did not differ significantly in comparison with spring. Regarding specific months among seasons, June was attained the highest significant ( $P<0.05$ ) AST and ALT activity, in spite of lowest significant ( $P<0.05$ ) AST during April and ALT during March, but another differences between months were not significant. Table 1 shows the mean of testosterone levels throughout the study, the ANOVA measurements on data normally distributed and showed an effect of different season on testosterone level for entire of the year ( $P<0.05$ ). The autumn season of present experiment was observed a rising significance [(8.85 $\pm$ 0.08) ng/mL] ( $P<0.05$ ), and a decreased significance ( $P<0.05$ ) was at the beginning of summer [(5.89 $\pm$ 0.06) (ng/mL), spring [(3.07 $\pm$ 0.04) ng/mL] and winter [(2.11 $\pm$ 0.08) ng/mL]. On monthly basis, testosterone concentrations (Table 1) in September, November, October August, June and July were greater significant ( $P<0.05$ ) than April, March, May, December, January and February. In addition, the differences significant ( $P<0.05$ ) between months of autumn compare with those of summer, and among March and April compare with December, January and February (Table 1).

## 4. Discussion

The result of present study revealed that the highest values of AST and ALT were calculated in summer season, and these results attribute to high environmental temperatures which affect not only performance parameters, but also require various physiological function[6–8]. However, Hussain *et al*[24] recorded that an increase

concentration and activity of AST in local goats was in winter, and serum ALT value was found to be increased during heat stress in goats[25]. Juma[26] arrived at the increased activity of the mentioned enzyme in the seminal plasma of bull in summer but no significant changes were observed in AST level in goats during heat stress[25–27]. The values of AST and ALT observed in present study are comparable with the values reported by Khawaskar *et al*[28] in semen of buffaloes and are higher than the values mentioned by Khawaskar *et al*[28] but are lower than the value of AST reported by Shukla *et al*[29]. The activities of ALT, AST and ALP significantly are elevated ( $P<0.05$ ) during wet summer season compared to other seasons (Vijay *et al* and Pandey *et al*[30,31] in cattle and Khawaskar *et al*[28] in buffaloes). The high temperature of the environment plays a role in the increase of cortisone in the circulatory system resulting in an increase in ALT/ALP enzymes due to the effect of oxidation on the liver tissue and the shale[32]. Marai and Habeeb[33] pointed out that the warming of the atmosphere leads to hormonal and enzymatic changes, but Juma and Al-Kassab[34] found the highest ( $P<0.01$ ) activity of ALT in ram semen was attained in spring, and effect of high temperature caused lowers ALP and lactic dehydrogease activity[35,36]. Decrease in these enzymes during heat stress is due to decrease in thyroid activity during heat stress[36]. Serum level of AST and ALT is helpful in diagnosis of welfare of animals. Ocak *et al*[27] and Sharma and Kataria[25] revealed that no significant changes were observed in AST level in goats during heat stress, Souhayla *et al*[37] reported that transaminase activity in the seminal plasma of bull semen was the highest in the poor ejaculates in comparison with good ejaculates in January in comparison with other months, and the same thing for good aculeate, the value was significant ( $P<0.05$ ) high in May in comparison with other months. In the present study, monthly variation in ALT and AST was observed, and these results are similar to those of Mohammed and Darren[38]. Mean of testosterone levels throughout the study showed an effect of the season on testosterone level for entire of the year. The autumn season of present experiment showed a rising significance of the testosterone level and the decrease significantly ( $P<0.05$ ) at the beginning of summer, spring and winter might attribute to the congenial weather condition which favored the testosterone activity[39]. In addition, results repeated that the differences significantly ( $P<0.05$ ) between summer, winter and spring season were similar to those of Sogorescu *et al*[40] for buck, but Okab[41] found blood plasma testosterone was observed in NZW rabbits in Egypt with higher in summer while with a decrease during spring, similarly[42–44]. Regarding testosterone concentrations in September, November, October, August, June and July were greater significant than April, March, May, December, January and July. Ram blood testosterone levels vary according to breed, nutrition level, season and age[45,46]. Sogorescu *et al*[40] reported that the highest levels of testosterone were found between August and September. In conclusion, the activities of both AST and ALT enzymes are highest during summer but testosterone is highest in autumn. On monthly basis, June showed the highest enzymes but November showed the highest testosterone hormone.

## Conflict of interest statement

Authors declare that there is no conflict of interest.

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