Seasonal effect on the scrotal circumference, concentration of testosterone and some biochemical parameters in Nuaimie breed rams in Salah-din province

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Abstract

The current study aimed to determine the effect of the seasons on the scrotal circumference, serum concentration of testosterone and some of the chemical parameters in Nuaimie breed rams in Salah-din province. Thirteen Nuaimie breed rams were used in this study. They were 3-4 years old and weighed 45-55 kg reared in a herd of sheep in Tikrit for the period from 15-8-2016 to 15-7-2017. Results showed that the scrotal circumference increased significantly (P≤0.05) during autumn (30.58±0.54 cm) and reaches lowest value during summer (29±0.32 cm). Highest concentration (P≤0.05) of testosterone was recorded in winter and autumn and lowest concentration in the spring and summer. The chemical analysis of ram blood serum revealed that the season has an effect on the value of triglyceride, glucose and cholesterol and the significant increase were recorded in autumn and winter. In conclusion, the reproduction in Nuaimie breed rams is characterized by distinct seasonal variations in scrotal circumference and blood testosterone concentration and that the highest measurement of these parameters are in autumn season. The season has a clear effect on the levels of triglycerides, glucose and cholesterol in ram's blood serum, which may give a good indication of reproductive performance in rams.

Key words: Seasonal effect, testosterone, scrotum circumference, reproduction.

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تأثير الموسم على محيط كيس الصفن وتركيز هرمون التستوستيرون وبعض المعايير الكيميائية للكلب في محافظة صلاح الدين

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الخلاصة

هدف الدراسة الحالية لمعرفة تأثير فصول السنة على محيط كيس الصفن وتركيز هرمون التستوستيرون وبعض المعايير الكيميائية في مصل دم الكلاب في محافظة صلاح الدين. استخدمت ثلاثة عشر كباً توارت اعمارها بين 3-4 سنوات يربون ضمن مزرعة في منطقة تكريت لمدة من 15-8-2016 إلى 15-7-2017. أظهرت النتائج أن الدراسة الحالية أن أعلى قياس لمحيط كيس الصفن قد سجل في فصل الخريف 0.54 ± 0.58 سم وفقر معنوي عن فصل الصيف 0.32 ± 0.29 سم. بينما كان أعلى تركيز لهرمون الاستروستيرون كان في فصل الشتاء وفقر معنوي (P<0.05) عن فصل الربيع والصيف. بلغ أعلى مستوى لمحيط كيس الصفن في فصل الخريف وفقر معنوي (0.5<P<0.05) عن فصل الربيع والصيف بينما كان أعلى تركيز للكولسترول في فصل الشتاء وفقر معنوي (P<0.05). تستنتج من الدراسة الحالية أن تأثير النمو بسمة موسمية في محيط الصفن وتركيز هرمون الاستروستيرون في الدم، وأن أعلى قياس سجل في موسم الخريف. كما أن الموسم تأثير واضح على مستوى كل من الدهون الثلاثية والكولسترول في مصل الدم في الكلاب، مما قد يعطي مؤشرًا جيدًا على الأداء التناسلي في الكلاب.

الكلمات المفتاحية: الموسمية، هرمون تستوستيرون، محيط كيس الصفن، الكولسترول.
Introduction

Unlike most domesticated animals, sheep are the best example of seasonal breeding animals (1). Lighting is the main factor responsible for seasonal changes in testicular activity by altering the secretion of LH and FSH from the pituitary gland and by its effect on the secretion of testosterone from the testicles (2), where testosterone is the main key to the spermatogenesis and thus the increase in germ cells number and testicular size in rams (3). While temperature, nutrition, mixed rearing, date of birth and period of lactation are modulator factors (4). In contrast, ewes that have no ovarian activity during non-breeding season, the sperm production and reproductive efficiency do not stop in rams, despite their obvious decline (1). The apparent effect of seasonality on the reproductive performance of rams is not sufficient to prevent the use of these rams during non-breeding season, despite the decrease in the quantity of sperm produced and the sexual desire of these males (6, 7). The size of the testis, usually estimated by measuring scrotal circumference, is one of the most important criteria used in evaluating the breeding soundness in rams (8). It has close relation to the concentration of sperm production from the testes (9). The season has a significant effect on the size of the testicles as the lack of daylight increases spermatogenesis and increases the circumference of the scrotum (10). As the largest diameter of the scrotal circumference can reach it in the fall and decrease by 2-3 cm in spring (7), so, the difference in testicular size during the annual breeding cycle of rams can be used as a measure of the degree of seasonal variation in different breeds (2). Many studies have revealed seasonal variations in the scrotal circumference, testosterone concentration and libido in many breeds of rams such as Persian Karakul (11), Texel Suffolk Dorset, Finnish Landrace (Finn), Suffolk and Scottish Blackface breeds (3), Droper (10), Chios and Serra rams (12) and Pampinta and Corriedale rams (13), Iranian Moghani (14), and Awaasi (15). While the effect of the season on reproductive performance is less in other breeds, which are raised in the tropical area such as Merino, Dorset, Romanov, Finn sheep and hair sheep (10) and Corriedale (16). In addition to the breed effect on the seasonal response of ram's production, individual differences in the same breed have a clear effect on this response (6), making it necessary to evaluate males to determine fertility for use in breeding programs throughout the year (17). Therefore, this study aimed to know the effect of the season on the concentration of testosterone and measuring the scrotal circumference, in addition to the concentration of some of the biochemical parameters in Nuaimie breed rams in Salah-din province.

Materials and Methods

- Animals: The study conducted in the period between 15/8/2016 to 15/7/2017 on 13 Nuaimie rams aged between 3-4 years and weighed 40-55 kg randomly selected from the breeding flock in the Tikrit city/ Salah-din province. Blood sample and clinical measurement were collected from each ram at each season of the year (autumn: September, October and November; winter: December, January and February; spring: March, April and may; summer: June, July and August) to conduct the effect of season on each parameter.

- Measurement of scrotal circumference and body condition score: Scrotal circumference assessment was conducted by using the flexible measurement tape, putting externally around the both testes in the widest area (6). The body conditions scored for each ram according to (18) by using scoring system 1 to 5 degrees (1 Emaciated, 2 thin, 3 Average, 4 fat, 5 Obese).

- Blood samples Collection and analysis: The blood samples were collected from the jugular vein of each ram throughout the year by using a dry test tube and put for
20 min. at room temperature, then blood samples were centrifuged 3000 rpm for 10 min. and the serum samples withdrawn and stored at -20 °C until the analysis.

The serum samples were analyzed at the AL-Maghrib laboratory for pathological analysis in Tikrit city. The testosterone concentration was measured using ELISA technique (Enzyme Linked Immuno-sorbent Assay) using a diagnostic commercial kit (Testosterone Enzyme Immunoassay Test Kit, Medix Biotech Inc., CA). Triglycerides, glucose, and cholesterol concentrations were quantitatively determined by colorimetric method using special kit provided by Biolabo, France.

- **Statistical analysis:** Statistical analysis of the data was obtained by extracting the average and standard error and comparing the averages in the ANOVA method. In order to obtain the highest mean, Duncan's multiple range tests were used using the statistical program (Sigma stat, Jandel scientific software V3.1 Inc.). Richmond, CA, USA, 2004.

### Results and Discussions

The results of the current study, as shown in Table (1), showed that the total measurement of the scrotal circumference during the period of study in the Nuaimie breed rams was (29.833 ± 0.242 cm). It is similar to what was recorded in Zandi rams (30.43±0.6 cm) in Iran (19), while it was lower than that recorded in other breeds such as Dorper rams (34.8±2.8 cm) in Hungary (10), Afshari and Shall breed (33.68±1.3 cm and 35.45±0.9 cm, respectively) in Iran (19), Awaasi (32.5±0.3 cm) in Jordan (6) and Karakul rams (32.0±1.2 cm) in Iran (11) and this difference illustrate the effect of breed on the measurement of the scrotal circumference in the rams (17).

**Table (1) Total the scrotal circumference, body condition score and concentration of Testosterone, Triglyceride, Glucose and cholesterol in Nuaimie ram in Iraq (mean ± SD)**

| Value | Scrotal circumference (cm) | BCS | Testosterone concentration (mg/ml) | Triglyceride (mg/ml) | Glucose (mg/ml) | Cholesterol (mg/ml) |
|-------|---------------------------|-----|-----------------------------------|---------------------|----------------|--------------------|
| Mean  | 29.833 ± 0.242            | 3.385 ± 0.068 | 7.065 ± 0.399 | 102.35 ± 8.429 | 55.42 ± 3.474 | 53.37 ± 2.836 |
| S/E   |                          |       |                                   |                     |                |                    |

The results of the current study showed that the season had a clear effect (p≤0.05) on the measurement of the scrotal circumference (Fig. 1), the highest rate recorded in the autumn and winter, while the lowest recorded in the summer and spring, similar observations were published by (10) in Dorper rams, (11) in Iranian Karakul rams, (17) in Awassi rams and (20) in Texel, Suffolk and Ile-de-France rams, in another study (21) the highest measure of the scrotum circumference in Brazilian Suffolk rams was recorded in spring and summer, This difference could be linked to the effect of climate and region (4).

![Figure (1): Seasonal effect on the scrotal circumference in Nuaimie rams](image)
The results of the current study showed (Table 1) that the body condition scores of the Nuaimei rams during the study period was 3.385 ± 0.068. This corresponds to (22) in Nuaimei rams (3.50±0.15) in Iraq. As for the effect of the season, the present study agreed with the findings of (23, 11), that there is no effect of the season on the degree of body condition score (Fig. 2). Even though the significant correlation between the body condition score and testicular circumference (6), the results of the present study showed no significant effect of the season on the body condition score, although its effect on scrotal circumference, this indicates that the measurement of the scrotum circumference is affected directly by light rather than by the effect of feeding (20).

![Graph](image)

Figure (2): Seasonal effect on body condition score for Nuaimei rams

The results of the current study, as shown in Table 1, demonstrated that the testosterone concentration in Nuaimei rams was (7.065±0.399 ng/ml), which is similar to what was recorded by (19) in al-Zandi rams (7.01±0.5 ng/ml) but it was higher than recorded by (11) in the Iranian Karakul rams (6.0±0.7 ng/ml), and by (19) in Afshari and Shall breed (6.4±0.81 and 5.59±0.76 ng/ml respectively) as well as (17) in Chios and Daglic breed (4.2±0.2 ng/ml and 3.7±0.2 ng/ml respectively). This indicates that despite the small scrotum circumference in Nuaimei rams compared to other breeds, the testosterone concentration was higher than the rest of the breeds and this describes that the secretion of testosterone is not directly related with the size of the testes (20), this illustrates the increase in the reproductive efficiency of Nuaimei ram, as high testosterone is a clear pointer to increased libido as well as an increase in sperm production and vitality in rams, (3, 20). The current study revealed significant difference in the testosterone concentration among different season, the highest value was recorded in winter and autumn and lowest in spring and summer (Fig. 3). Similar result was recorded in rams of several other breeds in many studies (2, 3, 17, 20). (11), (19) and (24) reported highest concentration of testosterone in summer and autumn while the lowest in winter and spring. These differences among studies may be due to differences between breeds and different latitude between studies involved (4, 17 and 21).
The results of the current study, as shown in Table 1, the concentration of triglycerides in the blood serum of Nuaimei rams was (102.35±8.42 mg/ml), which is lower than that obtained by (25) (188.31±7.05 mg/ml). Season had clear effect (P<0.05) on the triglyceride concentration in Nuaimei rams (Table 2), the concentration was higher in the autumn (190.7±7.46 mg/dl) than in the other season of year (73.1±3.585, 74.9±1.663 and 70.7±1.674 mg/dl for winter, spring and summer, respectively). Same result was submitted by (27) in Sakiz-Awassi crossbreed sheep.

### Table 2: Effect of different season on the blood concentrations of triglyceride, glucose and cholesterol in Nuaimei rams (mean ± SD).

| Parameters | Triglyceride (mg/ml) | Glucose (mg/ml) | Cholesterol (mg/ml) |
|------------|----------------------|----------------|---------------------|
| Autumn     | 190.7±7.46 a         | 68.7±2.381 a   | 65.3±1.886 a        |
| Winter     | 73.1±3.585 b         | 81.8±3.753 a   | 57.8±5.688 a        |
| Spring     | 74.9±1.663 b         | 34.8±1.652 b   | 32.5±2.242 b        |
| Summer     | 70.7±1.674 b         | 36.4±1.529 b   | 57.9±5.46 a         |

Different letters between raw mean significant differences at (P<0.05)

Glucose concentration during the experimental period was 55.42±3.47 mg/ml. This result was compatible with (27) in Yankasa sheep (54.05 ±1.88 mg/ml) while it was higher than (28) in Awassi rams (46.12±1.01 mg/ml) and (27) in Ouda and Balami rams (45.04±1.09 and 37.83±2.1 mg/ml respectively), this difference may be due to breed effect (29) or the nature of nutrition and management (30). Seasonal fluctuations of glucose concentration in rams were observed (table 2), the highest concentration was recorded in winter and autumn (81.8±3.753 and 68.7±2.381 mg/ml respectively) and significantly (P<0.05) decreased in spring and summer (34.8±1.652 and 36.4±1.529 mg/ml respectively), similar results were observed by (31) and this rise in glucose level in winter may be related to inhibition of insulin secretion due to the low nutrition because cold stress (32). The findings of the present study showed (Table 1) that the concentration of cholesterol (53.37±2.836 mg/ml) was higher than recorded by (27, 33) in Chios, Daglic and Yankasa rams (35.9, 32.6 and 45.04 mg/ml respectively), while it was lower than recorded by (27) in Ouda rams (63.06 mg/ml). This variation may be due to differences in breed (33). On the other hand, significant effect of season on the cholesterol concentration in Nuaimei rams was observed in the current study (Table 2). The highest concentration was recorded in autumn (65.3±1.886 mg/ml) and the lowest concentration in the spring (32.5±2.242 mg/ml). This result was compatible to the findings of (26, 30) who found clear variation in cholesterol concentration through different seasons in Sakiz-Awassi crossbreed sheep. The reason of this decrease in cholesterol concentration in the spring may be due to dilution as a result to the increase in total body water or to the decrease in acetate concentration, which is the primary precursor for the synthesis of cholesterol (34). We conclude from the current study that although Nuaimei rams have reproductive capacity throughout the year, their reproduction is characterized by distinct seasonal variations in scrotal circumference and blood testosterone concentration and that the highest measurement of these parameters are in autumn season. The season has a clear effect on the levels of triglycerides, glucose and cholesterol in rams blood serum, which were higher in the breeding season (autumn) and less in non-breeding season (spring), which may give a good indication of reproductive performance in rams.

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