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Effect D-Aspartic amino acid injection on some characteristics of semen in Shami Goats

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***Abstract*---**This study was conducted with the aim of showing the effect of injecting D-Aspartic acid on characteristics of semen, for Period 15/7/2021 until 15/10/2021 in the animal farm/ Ruminant Hall of Animal Production Department /College of Agriculture / University of Diyala. This study include 12 male of Shami goat aged (1-1.3) years, with an average weight (35-38) kg, were divided randomly to four equal groups (3/ male / group): the first was a control group (without injection) (T1), while the second (T2), third (T3) and fourth (T4) groups were injected intramuscular with D- Aspartic Acid in concentration 125, 250 and 375 mg /48 hour. Semen was collected by artificial vagina ،three times during the study. The results showed that the injection of D-Aspartic acid led to highly significant increase ($p<0.01$) in individual motility, Mass motility, live spermatozoa percentage, Dead spermatozoa ratio, sperm concentration, abnormal spermatozoa percentage. There were significant effects ($p<0.05$) for injection on the Mass motility of the sperm, where T4 and T3 were superior on T1 and T2. A highly significant effect was also found ($p<0.05$) for injection in the individual motility where T4 was superior on T1 and T2 and T3 and T2 were superior on T1. There was a significant effect of the injection in concentration, where T4 was followed by T3 then T2 and finally T1. The percentage of live spermatozoa was superior for T3 and T4 on T1 and T2 and T2 was superior on T1. There was a high significant effect in Dead spermatozoa so T4 and T3 were superior on T1 and T2, T2 was superior on T1 with the lowest percentage of Dead spermatozoa. There were also significant effects of injection in the percentage of abnormal form sperm, as T4 was superior on T1 and T2 and T3 and T2 were superior on T1 with the lowest percentage of abnormal spermatozoa.

Keywords---semen qualities, D-Aspartic acid, Shami goat.

Introduction

Livestock in Iraq occupies a prominent place in the national economic structure, especially the agricultural economy, because its products of red and white meat, milk and eggs are the main source of animal protein which is necessary for human food (Al-Qass and Faiq,1982). Goats are animals that are raised for multiple purposes, as they produce meat and milk in conditions that other farm animals cannot produce with the same efficiency (Mlambo and Mapiye,2015). Goat's milk is characterize by its medical properties and has recently gained importance in human health because it is easily digestible and close to the components of mother's milk, so the demand for it and its dairy products is expected to increase in the coming years (Elbehri, 2015). The shami or Damascus goat) is one of the important breeds of goats that originated in the Levant (Al-Qass et al., 1993). Its importance is due to its high efficiency in the production of milk and meat, with a high percentage of twins reaching to 75% in the mating season (Harba, 2020). In order to male reproductive process to continue with a high efficiency, the hormonal relationship must be maintained at high levels represented by the melatonin hormone, which is secreted from the pineal gland, GnRH hormone from the hypothalamus, SSH and ICSH from the pituitary gland, and testosterone from the testis, as they have an important role in the perpetuation and functioning of the sexual glands and the sperm formation and perpetuation of sexual behavior (Darbandi et al,2018). Since the Shami goat breed is one of the seasonal breeds in Iraq (Tallak, 2019), many studies have been conducted for the purpose of improving the seminal fluid quality of Shami goats male during the period of sexual inactivity through treatment with some hormones such as GnRH, hCG, Kisspeptin (Al-Amri, 2015) and luteinizing hormone. eCG (Al-Mahdawi, 2019) and eCG and hCG (Abdel Manhal, 2021) or treatment with vitamins such as vitamin C (Ishaq et al.,2005) or treatment with amino acids such as tryptophan (Mahdi, 2021).and Vitamin C has been used to improve the traits of the semen for rams(Al-Saab،2015)

D-Aspartic acid is an amino acid that is found in the nervous tissue and endocrine gland of both vertebrates and invertebrates (D'Aniello et al., 1998) and in high levels in the testicles (Di Fiore et al., 2014). It has an important role in the synthesis and secretion of endocrine hormones such as GnRH, SSH and ICSH. It regulates testosterone secretion (D'Aniello et al., 2000a) and spermatogenesis (Di Fiore et al., 2016). Most recent studies have been limited to testing the role of D-Aspartic acid in stimulating the secretion of GnRH, LH, FSH and testosterone in several animals, We believe that there is no study dealing with the role of D-Aspartic acid in improving the seminal fluid characteristics of Shami goats male during the period of sexual inactivity, therefore, this study was conducted to find out the effect of injecting different concentrations of D-Aspartic acid in the fertility of Shami goats by measuring its effect changes in semen characteristics.

Materials and Methods

This study was conducted in the field of the College of Agriculture / University of Diyala, 12 sexually mature Shami goats were used in this experiment, their ages ranged between (1-1.3) years and weights ranged between (35-38) kg. For the purpose of studying the effect of amino acid injection on some characteristics of semen in Shami goats in the summer, the animals were injected with acid and semen collected from male goats by using the artificial vagina of sheep and goats for once a month from each animal in the presence of a female in estrus injected with estradiol By (2.5) mg 36 hours before the collection process. The animals were divided into 12 male Shami goats with four treatments:-

- The first treatment, T1: the control treatment
- The second treatment T2: injection of D-Aspartic acid at a dose of 125 mg
- The third treatment T3: injection of D-Aspartic acid at a dose of 250 mg
- Fourth treatment T4: injection of D-Aspartic acid at a dose of 375 mg

The Mass motility of the sperms was assessed as reported by Blom (1946), and the individual motility was estimated as reported by Chemineau et al. (1991), the sperm concentration was calculated using an erythrocyte counting chip and prepared a dilution solution of 0.9% Sodium chloride and 0.01% mercury chloride and 2 g/l eosin dye (dissolved in water) to distinguish sperm under the microscope, so the dilution ratio was 1:200, and the live and Dead spermatozoa were calculated based on what was stated by Chemineau et al., (1991), the distorted sperm were calculated. According to the method of Chemineau et al., (1991). The statistical program CRD was used in the statistical analysis of the experiment data according to a design

- $Yijk= \mu + ai + eijk$
- Yijk = View value k of the transaction.
- μ = general mean of the experiment
- ai = main effect of the D-Aspartic acid
- eijk = the value of the experimental error of observation, which is distributed normally and randomly independently with mean equal to zero and variance 6e2.

Significant differences between means were compared using Duncan's polynomial test (Duncan, 1955).

Results and Discussion

The effect of D-Aspartic amino acid injection on individual and Mass activityand concentration of sperm

Table-1 showed the effects of D-Aspartic amino acid injections on the Mass motility of the sperm There were significant effects ($p<0.05$), where T4 and T3 were superior on T1 and T2 (71.77±2.07,79.55±3.31,85.11±2.68,86.11 ±2.38) %, respectively. A highly significant effect was also found ($p<0.01$) for injection in the individual motility where T4 was superior on T1 and T2 (72.00±1.25,78.66

±2.55,82.33 ±2.40,84.66±2.74) % respectively and T3 and T2 were superior on T1. There is a significant effect of the injection in concentration, where T4 was followed by T3 then T2 and finally T1(1.97±0.16,2.64±0.19,43.3±0.19,3.83±0.26)×10 ^{9} respectively.

Table 1
The effect of D-Aspartic amino acid injection on individual ، Mass activityand concentration of sperm (mean± SE)

Treatment	Individual movement %	Mass activity %	Spermatozoa concentration (ml ×109)
T1	1.25±72.00 c	2.07±71.77 c	0.16±1.97 d
T2	2.55±78.66 b	3.31±79.55 b	0.19±2.64 c
T3	2.40±82.33 ab	2.68±85.11 a	0.19±3.43 b
T4	2.74±84.66 a	2.38±86.11 a	0.26±3.83 a

Different letters in column indicate significant differences (a, b, c: P≤0.05)

The reason for increasing Mass motility of injection treatments may be due to the high level of testosterone to change the pattern of hormone secretion SSH and ISCH as a result of D-Aspartic acid injection (D'Aniello et al.; 2000a), with an increase in testosterone, the secretions of the accessory glands of energy sources will increase (Cevik et al., 2007), such as fructose and sorbitol, which provide the sperm with the energy needed for it's movement (Senger, 2003) this result agreed with (Raspa et al 2018) in the mouse. The increasing individual motility may be due to the high concentration of D-Aspartic acid in the epididymis (Daniello et al.; 2000a) which promotes sperm maturation and motility (Cornwall, 2009) by increasing energy sources (ATP) and increasing calcium influx (ca^{+2}) which increase tail motility of sperm (Barbato et al., 2017; Rodríguez-Miranda et al, 2008) also works on the activation of cAMP in Leydig cells that supplies sperm with energy (Valenti et al., 1999) as there positive correlation between mass activity and individual motility (Khalil،2018) and this result agreed with (Macchia et al., 2010) in rabbits male. The reason for the significant increase in the concentration of sperms may be due to the increase in the concentration of D-Aspartic acid in the seminiferous tubules of the testicles, which leads to an increase in the divisions of spermatogenetic cells (Spermatogonia) (Hasegawa et al., 2013). D-Aspartic acid activates AKT and ERK pathways within spermatogenetic cells that serve to provide proteins needed for spermatogenesis (Castoria et al., 2001) this result agreed with (D'Aniello et al, 2012) in men.

The effect of D-Aspartic amino acid injection on Live, Dead and Abnormal spermatozoa ration

Table-2 showed the effects of D-Aspartic amino acid injections on the percentage of live spermatozoa, T3 and T4 were superior on T1 and T2 (69.56±1.42,76.45±1.81 ,83.56±1.89,85.23±2.38) and T2 was superior on T1.

There is a high significant effect in Dead spermatozoa, T4 and T3 were superior on T1 and T2, T2 was superior on T1 with the lowest percentage of Dead spermatozoa (30.44±1.42,23.55±1.81,16.44±1.89 ,14.77±2.38(%) respectively. There was also significant effects of injection in the percentage of abnormal spermatozoa, T4 was superior on T1, T2 and T3 and T2 were superior on T1 with the lowest percentage of abnormal spermatozoa (18.55±1.02,11.77±1.11,10.00±0.64,8.77±0.95) % respectively.

Table 2
The effect of D-Aspartic amino acid injection on Live, Dead and Abnormal spermatozoa ration (mean± SE)

Treatment	abnormal spermatozoa%	Dead spermatozoa%	Live spermatozoa %
T1	1.02±18.55 c	1.42±30.44 c	1.42±69.56 c
T2	1.11±11.77 b	1.81±23.55 b	1.81±76.45 b
T3	0.64±10.00 ab	1.89±16.44 a	1.89±83.56 a
T4	0.95±8.77 a	2.38±14.77 a	2.38±85.23 a

Different letters in column indicate significant differences (a, b, c: P≤0.05)

The reason for increasing in the live spermatozoa percentage of the treatments may be due to increase in the number of Sertoli cells as a result of D-Aspartic acid injection, as these cells nourish the sperm by secreting a fluid rich in bicarbonate and potassium with the production of testosterone-binding protein (ABP) (Gado, 1996), which leads to an increase in the concentration of testosterone in the seminiferous tubules, which was reflected in the high live spermatozoa percentage (Al-Mahdawi, 2019), this result agreed with (Macchia et al., 2010) in rabbits male. The reason for the decrease in Dead spermatozoa may be due to the effect of D-Aspartic acid injection, which has an important role in the efficacy of ICSH and SSH hormones, which have an important role in improving the live spermatozoa percentage through their effect on Leydig and Sertoli cells, which led to an increase in the live spermatozoa percentage (Brunet et al., 2012). The reason for the low percentage of abnormal spermatozoa may be due to the injection of D-Aspartic acid, which raises testosterone in the seminiferous tubules and epididymis (Falvo et al., 2016), which is important in maintaining division of spermatogenic cells from the primary spermatocyte stage to spermatozoa (Sakai et al., 1998; Raucci and Di Fiore, 2009), which preserves the normal shape of sperm, raises their ratio and reduces the percentage of abnormal spermatozoa (Raspa et al., 2018).

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