sciendo

Archiva Zootechnica 26:2, 69-82, 2023
DOI: 10.2478/azibna-2023-0015

Effect of feeding dried olive (*Olea europaea*) leaves on the reproductive parameters of Awassi rams

Mohamad R. Al-Masri*, Moutaz Zarkawi, Mazen Alomar

*corresponding author: ascientific38@aec.org.sy

[1] *Division of Animal Production, Department of Agriculture, Atomic Energy Commission, P. O. Box 6091, Damascus, Syria. Tel: 2132580, Fax: 6112289,*

ABSTRACT

The aim of this study was to estimate the effect of feeding adult Awassi rams different ratio of dried olive leaves on the reproductive parameters during seven weeks during the breeding season. Three groups of Awassi rams were fed for twelve weeks either a conventional wheat straw-concentrate ration (Control) or experimental rations where 30% (G1) and 60% (G2) of the wheat straw were replaced with dried olive leaves. Animals were fed the same amounts of crude protein and metabolizable energy daily. Results indicated that with the exception of scrotal circumference, no significant ($p>0.05$) responses were noted for testosterone level, body weight, semen volume, sperm concentration or semen pH values between the tested groups. Analysis of motility characteristics of sperm by computer-assisted semen analysis (CASA) technique showed no significant differences ($p>0.05$) between the three groups in the percent motility, percent of sperm showing progressive motility, average path velocity, straight line velocity, curvilinear velocity, percent straightness and percent linearity. It could be concluded that feeding Awassi rams dried olive leaves even at the 60% level did not affect the reproductive characteristics of the Awassi rams and therefore could be used as supplement sources of feeds for small ruminants.

Keywords: feeding, olive leaves, reproduction, testosterone, semen quality

INTRODUCTION

Olive tree, typical of the Mediterranean vegetation, is well tailored to drought, poor soils, and resistant to salinity, where more than 70 % of world olive oil production comes from the Mediterranean region (Guerrero Maldonado et al. 2016). Moreover, olive oil industry produces large amounts of leaves, olive cake and water waste as "olive molasses" (Molina-Alcaide and

Yanez-Ruiz, 2008). Olive leaves represent around 10% of total weight arriving at olive extraction (Delgado et al. 1998). Air-drying for 7 days in the shade at room temperature did not have detrimental effects on the nutritional components of olive leaves and could represent a simple and low-cost procedure for their preservation (Martin-Garcia and Molina-Alcaide, 2008). An important alternative utilization of olive leaves would be as roughage feeds for ruminants (Paiva-Martins et al. 2009). Such form of nutrition could improve the economy and the efficiency of animal production (Molina-Alcaide and Nefzaoui, 1996). We previously noted that replacement of wheat straw by dried olive leaves in the rations of Awassi lambs had no effect on the daily weight gain and also on the feed conversion efficiency (FCE) (Alomar et al. 2022a). Moreover, substitution of wheat straw with dried olive leaves in rations of Awassi rams at levels up to 60% did not also affect FCE parameter and the performance of the tested animals (Alomar et al. 2022b).

It is well recognized that nutrition and reproduction have always been linked in that the reproductive success of an animal depends on its nutritional status (Brown, 1994; Robinson, 1996). In this respect, nutrient either in deficient or in higher amounts has been shown to be capable of altering reproduction, but the basic problem was always in the degree of the excess, deficiency or imbalance which was required to alter the reproduction status (Bindari et al. 2013). However, nutrition is considered to be an important element affecting seasonality of reproductive functions in small ruminants (Zarazaga et al. 2009). Zarkawi et al. (2003) reported that 50% of Damascus does fed lentil straw and concentrate plus *Sesbania aculeate* hay failed to kid, and they attributed this either to a failure to conceive or to an early embryonic mortality, whereas, partial substitution of *Atriplex lentiformis* (Al-Masri et al. 2007) and *Sesbania aculeate* (Zarkawi et al. 2005) with wheat straw showed no effects on the reproductive parameters of Damascus does and Awassi ewes, respectively.

It must be stressed out that several reproductive parameters may be directly influenced by nutrition. In both human and animals, insufficient supply in the diet may result in a negative effect with regard to spermatogenesis, sperm quality and male fertility (Salas-Huetos et al. 2017). Also, the scrotal circumference and testicular size are significantly affected by nutrition (Masters and Fels, 1984). Kheradm et al. (2006) confirmed that sperm production, as well as total number of spermatozoa per ejaculate increased by improved diet. Furthermore, increasing energy levels by 20% in diet has been indicated to get better sperm motility and velocity as well as mitochondrial membrane potential in rams (Selvaraju et al. 2012). Anyhow, sperm motility and semen quality assessments have long been reported as major criteria of semen analysis for predicting male fertility and is, therefore, emphasized heavily in fertility examination (Budworth et al. 1988; Mortimer, 1997). On the other hand, testosterone has a central role in controlling of

spermatogenesis, testicular state, and sexual behaviour in rams (Selvak kumar et al. 2017), and the level of testosterone secretion may be affected by nutrition regime (Walkden-Brown et al. 1994)

There is a dearth of information concerning the use of unconventional feeds as a feed supplement in diets and their effects on the reproductive performance of small ruminants. In a review paper, Bandari et al. (2013) reported the interaction between reproduction and nutrition affecting the reproductive status of animals. As we previously studied the effects of dried olive leaves feeding on the several nutritional parameters for both Syrian Awassi lambs and rams (Alomar et al. 2022a, b), we are interested to study the effects of such nutrition regime on the reproductive status of Awassi rams. For that, the main objectives of the current study were to determine the effect of feeding adult Awassi rams different ratio of dried olive leaves (30 % and 60 %) on the reproductive parameters during the breeding season.

MATERIALS AND METHODS

Site description, material tested and ethical approval

The present study was carried out at Der Al-Hajar Centre, about 33 km South-East of Damascus. This is dry area with an annual rainfall of about 140 mm. Olive *(Olea europea)* leaves as a by-product of olive oil extraction, were collected from four factories about 20 km north of Damascus and dried in the shade for seven days. The dried leaves were thoroughly mixed to be used later as feed supplement for the experimental Awassi rams. This study was approved by the Local Scientific and Ethical Committee of the Atomic Energy Commission of Syria, Damascus, Syria (permit number 36-Z/M, 2-2019).

Experimental animals, diets and management

At the start of the study, twelve adult Syrian Awassi rams aged between 2 and 3 years were randomly allocated to three similar groups in terms of age and initial body weight: the Control group (n=4; 71.3 ± 9.6 kg); the first experimental group (G1; n=4; 71.8 ± 4.7 kg); and the second experimental group (G2; n=4; 68.3 ± 6.1 kg). Rams were individually housed in metal pens. The Control group was fed a conventional wheat straw-concentrate ration containing 51% wheat straw and 49% concentrate, which contained 90.1 g/kg crude protein (CP) and 8.37 MJ/kg metabolizable energy (ME). For the two experimental groups G1 and G2, portions (30 and 60%, respectively) of the wheat straw were replaced by dried olive leaves. Level of concentrate in the experimental rations was adjusted, while the level of concentrate mixture was reduced and barley added to make all rations (Control and Experimental) isocaloric and isonitrogenous as recommended by Kirchgessner (1982) and

Friesecke (1984). Animals were fed the same amounts of CP (166 g) and ME (15.3 MJ) daily. However, in a previous study, Alomar et al. (2022b) gave the details of the nutritive components of the feed ingredients and the balanced rations for the three groups. The study lasted for twelve weeks during the breeding season (from June to August) (Zarkawi, 1997) under natural photoperiod and temperature conditions. Rams were fed the same rations for 5 weeks to allow them to adapt to the rations before the beginning of the 7-week experimental period. All the twelve rams were weighed at the beginning of the study and weekly during the experimental period at the same time in the morning before feeding.

Blood samples collection and testosterone levels determination

Blood samples (10 mL) were collected from the jugular vein of all experimental animals weekly at the same time using vacutainers. Serum was harvested by centrifugation of blood at 3000 rpm for 20 minutes and stored at -20°C until assayed using validated testosterone ELISA kits (Dia Metra S.r.I., SPELLO (PG), Italy). ELISA reader (Human Reader HS, Wiesbaden, Germany) was used.

Scrotal circumference measurement, semen collection and semen parameters analysis

During the 7-week experimental period, scrotal circumference (SC) was measured weekly at the largest part of scrotum by using a measuring tape. For semen collection, an electrical stimulation was applied using an electro-ejaculator (ElectroJac 5, Ideal Instruments, USA) administrating a series of 32 cycles pulses of short electrical stimuli with each cycle (two seconds on, then two seconds off) delivering a slightly higher intensity until semen production. The entire procedure was performed in approximately 2 min and repeated directly if the ram did not respond, and animals were held in a standing position during treatments. In this position, the penis of the rams was easily grasped and held into the end of calibrated centrifuge tube.

A total of 84 ejaculates were used in this study and the ejaculates were assessed for semen volume, sperm concentration, semen pH and motility characteristics. The total volume of the ejaculate was measured using a calibrated test tube, the pH values were measured using a pH-meter (Bibby Scientific, Model 3505, United Kingdom). Sperm concentration was assessed using a haemocytometer counting chamber (Neubauer Improved Marienfeld, Germany) according to the method of the World Health Organization (1992).

The motility characteristics of the sperm were immediately assessed after semen collection by CASA technique, using the Hamilton-Thorne motility analyzer (HTM version 12.3, USA). The analyzer setting is displayed in Table 1.

Table 1. Setting for the Hamilton Thorne IVOS animal breeders software (version 12.1) used to evaluate ram sperm kinematics

Parameter	Settings
Frame rate (Hz)	60
Frames acquired (no)	30
Minimum contrast	60
Minimum cell size (pixels)	5
Low average path velocity (VAP) cut off	21.99
Low straight line velocity (VSL) cut off	6
Non-motile head size (pixels)	5
Non-motile head intensity	55
Static size limit (min/max)	0.60/8
Static intensity limit (min/max)	0.20/1.50

Prior to semen analysis, each sample was diluted to approximately 25 x 106 spermatozoa/ml in Tris-Fructose basic solution (Tris 36.3 g/l, citric acid 19.9 g/l, Fructose 5 g/l; 300 mOsm/kg; pH 6.8). The motility characteristics included in the analysis were: the percent motility, curvilinear velocity (VCL, μm/s), average path velocity (VAP, μm/s), straight line velocity (VSL, μm/s), percent linearity (LIN, %), percent straightness (STR, %) and the percent of sperm showing progressive motility (PMOT %: VAP ≥ 75 μm/s and STR ≥ 80 %). More details about these parameters were reported by Verstegen et al. (2002).

Statistical analysis

A 7 x 3 factorial-design was used in this experiment, with two fixed factors: (1) housing period (seven weeks); (2) ration type (Control, G1 and G2). Results were subjected to analysis of variance (ANOVA) using a Statview-IV program to test the effects of housing period and ration type treatment on the studied parameters. Means were separated using the Fisher's least significant difference test at the 95% confidence level.

RESULTS

Table 2 shows the effect of partial replacement of wheat straw with olive leaves (G1; 30 % and G2; 60 %) in rations of Awassi rams on body weight, testosterone level, scrotal circumference, semen volume, sperm concentration and semen pH.

Table 2. Effect partial replacement of wheat straw with olive leaves (G1; 30 % and G2; 60 %) in rations of Awassi rams on body weight, testosterone level, circumference, semen volume, sperm concentration and semen pH values of collected semen during seven housing periods (W)

	Weight (Kg)	Testosterone (nmol/L)	Circumference (cm)	Volume (ml)	Concentration (x109/ml)	pH
Period (P) (pooled, n=12)						
W1	71.8^{a}	7.35^{a}	31.4^{bc}	1.49^{a}	1.70^{a}	6.33^{a}
W2	72.7^{a}	7.84^{a}	31.1^{c}	1.36^{a}	1.53^{a}	6.07^{b}
W3	73.6^{a}	6.95^{a}	31.9^{abc}	1.37^{a}	1.42^{a}	6.28^{ab}
W4	73.7^{a}	6.64^{a}	32.4^{ab}	1.42^{a}	1.53^{a}	6.18^{ab}
W5	73.6^{a}	6.88^{a}	32.5^{ab}	1.41^{a}	1.49^{a}	6.13^{ab}
W6	73.7^{a}	7.55^{a}	32.4^{ab}	1.46^{a}	1.53^{a}	6.03^{b}
W7	75.7^{a}	8.87^{a}	32.7^{a}	1.44^{a}	1.53^{a}	6.03^{b}
SEM	1.56	1.68	0.43	0.10	0.11	0.09
Ration (R) (pooled, n=28)						
Control	73.5^{a}	6.96^{a}	32.5^{a}	1.41^{a}	1.49^{a}	6.14^{a}
G1 (30 %)	75.6^{a}	6.85^{a}	31.1^{b}	1.44^{a}	1.56^{a}	6.10^{a}
G2 (60 %)	71.6^{a}	8.43^{a}	32.5^{a}	1.41^{a}	1.55^{a}	6.21^{a}
SEM	1.09	1.08	0.253	0.061	0.070	0.060
			Interaction (PxR)			
p-value	>0.9999	>0.9999	0.9537	0.9980	0.9961	0.2523

^{a,b,c} Means in the same column with different superscripts are different at $p<0.05$. SEM: Standard error of the means.

Animals live-body weights during the experimental period were not significantly different ($p>0.05$) between the two experimental groups fed on dried olive leaves and the Control group and amounted in average to 73.6 kg. Serum testosterone levels during the experimental period were not significantly different ($p>0.05$) between the two experimental groups fed on dried olive leaves and the Control group being 6.96, 6.85 and 8.43 nmolL^{-1} for Control, G1 (30%) and G2 (60), respectively, with an overall mean reaching 7.41 nmol L^{-1} on average and for all the tested animals, the SC significantly ($p<0.05$) increased from 31.4 Cm to 32.7 Cm at the end of the experiment. The G1 group significantly differed ($p<0.05$) from both G2 and the Control group. However, no significant effect ($p>0.05$) was noted for the interaction between the housing period and the type of the ration. The average semen pH for the 12 rams ranged between 6.03 and 6.33, with significant differences ($p<0.05$) between the seven weeks of the experimental housing period. Anyhow, the semen characteristics did not show significant differences ($p>0.05$) for the semen volume, sperm concentration and also for the semen pH values between the three experimental groups.

The comparison of sperm motility characteristics between the three groups analyzed by CASA system is presented in Table 3.

Table 3. Effect partial replacement of wheat straw with olive leaves (G1; 30 % and G2; 60 %) in rations of Awassi rams on CASA motility characteristics during seven housing periods (W)

	MOT (%)	PMOT (%)	VAP (µm/s)	VSL (µm/s)	VCL (µm/s)	STR (%)	LIN (%)
Period (P) (pooled, n=12)							
W1	80.6^{ab}	26.0^{a}	135.6^{a}	100.4^{a}	226.6^{a}	69.1^{a}	42.9^{a}
W2	82.2^{a}	21.7^{b}	122.1^{bc}	87.3^{bc}	210.8^{b}	68.5^{a}	40.9^{a}
W3	83.0^{a}	21.0^{b}	115.4^{c}	82.5^{c}	184.2^{d}	68.5^{a}	42.1^{a}
W4	78.3^{abc}	21.7^{b}	118.2^{c}	85.8^{bc}	195.5^{d}	68.2^{a}	42.7^{a}
W5	77.1^{bc}	20.9^{b}	116.9^{c}	85.2^{bc}	194.1^{d}	69.1^{a}	43.5^{a}
W6	80.5^{ab}	19.1^{b}	120.4^{bc}	83.7^{c}	198.1^{cd}	66.9^{a}	41.9^{a}
W7	73.6^{c}	19.1^{b}	125.7^{b}	91.3^{b}	206.1^{bc}	68.1^{a}	42.6^{a}
SEM	1.56	0.83	2.47	2.35	3.53	1.16	1.06
Ration (R) (pooled, n=28)							
Control	78.9^{a}	21.5^{a}	120.3^{a}	87.6^{a}	202.1^{a}	68.8^{a}	42.4^{a}
G1 (30 %)	79.2^{a}	21.6^{a}	124.2^{a}	89.6^{a}	205.9^{a}	68.5^{a}	43.1^{a}
G2 (60 %)	79.9^{a}	20.9^{a}	121.6^{a}	86.8^{a}	202.9^{a}	67.8^{a}	41.6^{a}
SEM	1.16	0.74	1.96	1.84	3.12	0.75	0.69
Interaction (PxR)							
p-value	0.9708	0.8906	0.8245	0.6591	0.6426	0.9733	0.6799

^{a,b,c,d} Means in the same column with different superscripts are different at P<0.05. SEM: standard error of the means. MOT %: percent motility; PMOT %: percent of sperm showing progressive motility; VAP: average path velocity; VSL: straight line velocity; VCL: curvilinear velocity; LIN %: percent linearity, STR %: percent straightness.

Analysis of motility characteristics of sperm showed no significant differences (p>0.05) in the percent of sperm motility (MOT%), the percent of sperm showing progressive motility (PMOT), and velocity parameters VSL, VCL, VAP and the trajectory parameters STR % and LIN between the Control group and the experimental groups. Moreover, for all CASA motility parameters, no significant effects were noted for the interaction between the housing period and the type of the ration.

DISCUSSION

The present study is considered the first which clearly shows the effects of feeding dried olive leaves on the reproductive parameters of Awassi rams. Generally, nutrition is of primary importance in determining the reproductive status and semen quantity and quality (Robinson, 1996; Robinson et al. 2006; Martin et al. 2010; Clement et al. 2012). It must be emphasized that our

experiment was conducted under nutritionally controlled conditions and a period of 5 weeks feeding adaptation was applied before the beginning of the experimental period for 7 weeks. In fact, in mature rams there is 6 to 7 weeks delay in the response of spermatozoa numbers to diet, reflecting the time it takes for the development of spherical spermatids in the germinal epithelium to fully mature spermatozoa in the distal cauda epididymidis (Robinson, 1996). Thus, all the recorded values of the assessed reproductive parameters especially the semen parameters for the two experimental groups of this study reflect the effects of feeding dried olive leaves during the present experimental period.

At the start of this study, animals were normally growing because they were young adult rams; therefore, feeding was adjusted to allow appropriate development of the animals in the three groups. It must be emphasized that our experiment was conducted under nutritionally controlled conditions rather than in an extensive production system with no changes in the intake of ME and CP between the three groups. The absence of significant differences between the two experimental groups and the Control group concerning body weight indicates the no adverse effect of including dried olive leaves in Awassi rams' diets. In this respect and as in our previous work, we found no significant differences in the feed conversion efficiency between the Control and the same treated Awassi rams fed on dried olive leaves up to 60 % in the rations (Alomar et al. 2022b).

One of the most essential results of this study was that olive leaves' feeding has no effects on testosterone level. It is well known that hormonal responses play a role in mediating the physiological and behavioural processes that influence human and animal fertility (Norman and Smith, 1992). Testosterone secretion in small ruminants was driven by natural day length changes or artificial photoperiodic cycles (Langford et al. 1999). However, Syrian Awassi rams displayed a clear seasonality of testosterone levels where the highest level was recorded in June, while the lowest was noted in December (Alomar et al. 2016). In the present study, there were no significant differences ($p>0.05$) in mean serum testosterone level between the two experimental groups and the Control group being 6.96, 6.85 and 8.43 nmol L^{-1} for Control, G1 (30%) and G2 (60), respectively, with an overall mean reaching 7.41 ± 1.08 nmolL^{-1}, which is close to 9.46 nmol L^{-1} in Syrian Awassi rams at the age of 13 months using RIA technique (Zarkawi and Al-Daker, 2016).

Our present investigation was conducted during the breeding season of Awassi ram where testosterone has its highest secretion level. Since the plasma concentration of testosterone was considered as an indirect indicator of hypothalamo–pituitary activity, the findings of Perez-Clariget et al. (1998) indicated that the yearly variation in scrotal circumference and plasma testosterone concentrations in the Corriedale rams were not strictly

dependent on feed supply and that the hypothalamo–pituitary system operates independently of nutrition, at least during certain periods of the reproductive cycle. Moreover, it was suggested that the energy intake in diet probably had no affect either on ram spermatogenesis or on testosterone production directly, but may influence the reproductive parameters via an effect on gonadotrophin secretion (Brown, 1993).

Compared to the Control group, scrotal circumference did not differ from the G2 group and this probably related to the same balanced diet provided to the two groups. Despite G1 group was significantly different from the Control and G2 groups for this parameter, it was clear that the interaction between the period of the study and the ration did not differ. Anyhow, SC in rams how s a clear seasonal pattern and tends to parallel the seasonal changes in live weight and the hormonal profile (Perez-Clariget et al. 1998). Masters and Fels (1984) data showed that testicular size is controlled by nutrition, even to the extent that well fed rams in spring may have larger testes than poorly fed rams in autumns. It appears that nutrition-induced increase in testicular size is associated with either an increase in the size and length of the seminiferous tubules (Oldham et al. 1978) or an increase in the number of expanded tubules (Nolanet al. 1990).

No differences were noted for semen volume, sperm concentrations and semen pH between the two experimental groups and the Control group. The averages of both semen volume and sperm concentrations recorded in this study were within the ranges of Awassi breeds suggested by Kridi et al. (2006). According to Kheradmand et al. (2006), semen volume was not significantly affected by improved dietary intake above maintenance requirement, whereas the data of Oldham et al. (1978) showed that rams maintained on high plane of nutrition produced more sperm than those raised under deficient nutrition.

Computer-assisted sperm analysis (CASA) has been successfully used for over 30 years for routine and research purposes in human fertility centers and veterinary clinics (Agarwal et al. 1992; Verstegen et al. 2002; Amann and Waberski, 2014), allowing objective, simultaneous, reliable, and accurate analysis of semen motility parameters. We previously used this important system to analyse the effects of different agents and conditions on the motility status of Awassi rams (Alomar et al. 2018; Alomar, 2022). Motility is widely supposed to be one of the most important characteristics linked with the fertilizing ability of sperm. The measurement of sperm velocity parameters has been considered as an indicator of mitochondrial function in spermatozoa (Graham et al. 1984). It is noteworthy that some CASA variables are correlated with fertility and the combinations of several motility variable resulted in greater r2 values (0.68-0.98; Farrell et al. 1998). Thus, analyzing motility and velocity parameters generated by CASA technology in our study was useful to give a preliminary insight about the effects of the three experimental rations

on rams' fertility of the three groups. Smith et al. (1998) reported a significant correlation of CASA motility parameters and field fertility, while Robayo et al. (2008) have showed the relation between CASA motility patterns and ram sperm migration in ewe cervical mucus; and they reported that VCL and VAP were the initial sperm kinematic which positively correlated with the ability of sperms to migrate in ewe cervical mucus. In our study, the proportion of actively motile spermatozoa assessed by CASA system did not differ between the three experimental groups. Moreover, all the indicators characterizing velocity of spermatozoa such as VAP, VSL, VCL, and the trajectory of spermatozoa such as STR % and LIN % were not significantly different between the three groups.

Generally, the absence of significant responses on testosterone level, semen volume, sperm concentration, semen pH and motility score between the three groups most likely indicates that inclusion of dried olive leaves up to 60 % as a substitution of wheat straw in Awassi rams rations would have no adverse effects on the accessory sex glands. Anyhow, additional researches on the effect of dried olive leaves diets on fertility using evaluating techniques directly related to fertilization process such as IVF assays are needed to draw final conclusion.

CONCLUSION

Taken together, the data suggests that feeding Awassi rams different rations of dried olive leaves even at the 60% level did not change body weight and the most important reproductive characteristics. Our study has particular importance in that the Awassi is a breed of which the interaction between nutrition and reproduction has been rarely described. Finally, olive leaves could play a very important role as a source of feeds in the present and future development of extensive animal production system

ACKNOWLEDGEMENTS

The authors would like to thank the Director General and the Head of the Department of Agriculture of the Atomic Energy Commission of Syria for their encouragement.

REFERENCES

Agarwal, A., Ozturk, E., Loughlin, K.R., 1992. Comparison of semen analysis between the two Hamilton-Thorn semen analysers. Andrologia. 24, 327-328. DOI: 10.1111/j.1439-0272.1992.tb02663.x

Al-Masri, M.R., Zarkawi, M., Khalifa, K., 2007. Partial substitution of *Atriplex lentiformis* for wheat straw in the diet of Damascus does. Tropic Grasslands. 41, 301-307. Corpus ID: 73591117

Alomar, M., Soukouti, A., Alzoabi, M., Zarkawi, M., 2016.Testosterone and cortisol patterns and the effects of electro-ejaculation and copulation in Awassi rams. Archives Anim Breed. 59, 139-144.https://doi.org/10.5194/aab-59-139-2016, 2016

Alomar, M., Zarkawi, M., Alzoabi, M.A., 2018. Analysis of Awassi sperm motility in two media at different levels of temperature, pH and osmolality. Iranian J Appl Anim Sci. 8 (3), 167-172.http://www.scopus.com/inward/record.url?eid=2-s2.0-85052786541&partnerID=MN8TOARS

Alomar, M., 2022. Antioxidant potential of pentoxifylline on spermatozoa of small ruminants. Asian Pacific J Reprod. 11(3), 138-145.DOI: 10.4103/2305-0500.346092

Alomar, M., Al-Masri, M.R., Zarkawi, M., 2022a. Feed conversion efficiency of male and female Awassi lambs fed on dried olive *(Olea europaea)* leaves. Spanish J Agric Res. 20(2).e06SC02.https://doi.org/10.5424/sjar/2022202-18119

Alomar, M., Al-Masri, M.R., Zarkawi, M., 2022b. Effects of feeding dried olive *(Olea europaea)* leaves with wheat straw concentrate rations on feed conversion efficiency in Awassi rams. Tropic Grasslands-Forrajes Tropicales. 10(3), 297–301. DOI:10.17138/TGFT(10)297-301

Amann, R.P., Waberski, D., 2014. Computer-assisted sperm analysis (CASA): Capabilities and potential developments. Theriogenology. 81, 5-17.https://doi.org/10.1016/j.theriogenology.2013.09.004

Bindari, Y.R., Shrestha, S., Shrestha, N., Gaire, T.N., 2013. Effects of nutrition on reproduction- A review. Adv Appl Sci Res. 4(1), 421-429.www.pelagiaresearchlibrary.com

Brown, B.W., 1994. A review of nutritional influences on reproduction in boars, bulls and rams. Reprod Nutr Dev. 34, 89-114.DOI: 10.1051/rnd:19940201

Budworth, P.R., Amann, R.P., Chapman, P.L., 1988. Relationships between computerized measurements of motion of frozen-thawed bull spermatozoa and fertility. J Androl. 9,41-54. DOI: 10.1002/j.1939-4640.1988.tb01007.x

Clement, C., Witschi, U., Kreuzer, M., 2012. The potential influence of plant – based feed supplements on sperm quantity and quality in livestock: A review. Anim ReprodSci.132, 1-10.DOI: 10.1016/j.anireprosci.2012.04.002

Delgado Pertinez, M., Chesson, A., Provan, G.J., Garrido, A., Gomez-Cabrera, A.,1998. Effect of different drying systems for the conservation of olive leaves on their nutritive value for ruminants. Annales De Zootechnie. 47,

141-150.HAL Id: hal-00889721https://hal.archives-ouvertes.fr/hal-00889721

Farrell, P.B., Presicce, G.A., Brockett, C.C., Foote, R.H., 1998. Quantification of bull sperm characteristics measured by computer-assisted sperm analysis (CASA) and the relationship to fertility. Theriogenology. 49, 871-879.DOI: 10.1016/S0093-691X(98)00036-3

Friesecke, H., 1984. Handbuch der praktischen Fütterung. München (Germany): BLV- Verlagsgesellschaft mbH.

Graham, E.F., Schmehl, M.K.L., Deyo, R.C.M, 1984. Cryopreservation and fertility of fish, poultry and mammalian spermatozoa. In: Proc 10th NAAB Technical Conf Artificial insemination Reprod, 12-14 April 1984, Milwaukee, WI. National Assoc Anim Breeders, Columbia, MO, Pp 4-24.

Guerrero Maldonado, N., López, M.J., Caudullo, G., de Rigo, D., 2016. *Olea europaea* in Europe: distribution, habitat, usage and threats. In: San-Miguel-Ayanz, J., de Rigo, D., Caudullo, G., Houston Durrant, T., Mauri, A. (Eds.), European Atlas of Forest Tree Species. Publ. Off. EU, Luxembourg, pp. e01534b+ https://www.researchgate.net/publication/299470438

Kheradmand, A., Babaei, H., Batavani, R.A., 2006. Effect of improved diet on semen quality and scrotal circumference in the ram. Veterinarski Arhiv. 76 (4), 333-341.

Kirchgessner, M., 1982. Tierernährung, 5. Auflage. Frankfurt (Germany): DLG-Verlag.

Kridi, R.T., Abdullah, A.Y., Momani Shaker, M., 2006. Sexual performance and reproductive characteristics of young adult Awassi, Charollais-Awassi and Romanov-Awassi rams. Sheep Goat Res J. 21, 12-16.

Langford, G.A.,Sanford, L.M., Marcus, G.J., Shrestha, J.N.B., 1999. Seasonal cyclic pituitary and testicular activities in rams, Small Rumin Res. 33, 43-53.https://doi.org/10.1016/S0921-4488(98)00199-0

Martin-García, I., Molina-Alcaide, E., 2008.Effect of different drying procedures on the nutritive value of olive *(Olea europaea* var. europaea) leaves for ruminants. Anim Feed SciTechnol.142, 317-329.DOI: 10.1016/j.anifeedsci.2007.09.005

Martin, G.B., Blache, D., Miller, D.W., Vercoe, P.E., 2010. Interactions between nutrition and reproduction in the management of the mature ruminant. Animal. 4, 1214-1226.DOI: 10.1017/S1751731109991674

Masters, D.G., Fels, H.E., 1984. Seasonal changes in the testicular size of grazing rams. Proc Aust Soc Anim Prod.15, 444-447.

Molina-Alcaide, E., Nefzaoui, A., 1996. Recycling of olive oil by-products: possibilities of utilization in animal nutrition. Inter Biodeter Biodegr. 38, 227-235. https://doi.org/10.1016/S0964-8305(96)00055-8

Molina-Alcaide, E., Yanez-Ruiz, D.R.,2008. Potential use of olive by-product in ruminant feeding: A review. Anim Feed Sci Technol. 147, 247-264.https://doi.org/10.1016/j.anifeedsci.2007.09.021

Mortimer, S.T., 1997. A critical review of the physiological importance and analysis of sperm movement in mammals. Hum ReprodUpdate. 3, 403-439.DOI:10.1093/humupd/3.5.403

Nolan, C.J., Neuehdorff, D.A., Godfrey, R.W., Harms, P.G., Welsh, T.H., McArthur, N.H., Randel, R.D., 1990. Influence of dietary energy intake on prepubertal development of Brahman bulls. J Anim Sci. 64(4), 1087-1096.DOI: 10.2527/1990.6841087x

Norman, R.L., Smith, C.J., 1992. Restraint inhibits luteinizing hormone and testosterone secretion in intact male rhesus macaques: Effects of concurrent naloxone administration. Neuroendocrinology. 55, 405–415.DOI: 10.1159/000126151

Oldham, C.M., Adams, N.R., Gherardi, P.B., Lindsay, D.R., Mackintosh, J.B., 1978. The influence of level of feed intake on sperm producing capacity of testicular tissue in the ram. Aust J Agric Res. 29, 173-179.https://doi.org/10.1071/AR9780173

Paiva-Martins, F., Barbosa, S., Pinherio, V., Mourao, J.L., Outro-Monteiro, D., 2009. The effect of olive leaves supplementation on the feed digestibility, growth performances of pigs and quality of pork meat. Meat Sci. 82, 438-443.https://doi.org/10.1016/j.meatsci.2009.02.014

Perez-Clariget, R., Forsberg, M., Lopez, A., Castrillejo, A., 1998. Effect of nutrition on seasonal changes in scrotal circumference, testosterone and pituitary responsiveness to exogenous GnRH in Corriedale rams. Small Rumin Res. 29, 61-69.https://doi.org/10.1016/S0921-4488(97)00103-X

Robinson, J.J., 1996. Nutrition and reproduction. Anim Reprod Sci 42: 25-34.https://doi.org/10.1016/0378-4320(96)01526-6

Robinson, J.J., Ashworth C.J., Rooke J.A., Mitchell L.M., McEvoy T.G., 2006. Nutrition and fertility in ruminant livestock. Anim Feed Sci Technol. 126, 259-276.https://doi.org/10.1016/j.anifeedsci.2005.08.006

Robayo, I., Montenegro, V., Valdés, C., Cox, J.F., 2008. CASA assessment of kinematic parameters of ram spermatozoa and their relationship to migration efficiency in ruminant cervical mucus. Reprod Dom Anim. 43, 393–399.DOI: 10.1111/j.1439-0531.2007.00920.x

Salas-Huetos, A., Bullo, M., Salas-Salvado, J., 2017. Dietary patterns, foods and nutrients in male fertility parameters and fecundability: a systematic review of observational studies. Hum Reprod Update. 23, 371-389.DOI: 10.1093/humupd/dmx006

Selvak kumar, R., Sivakumar, T., Kumar, S.G., Anuradha, P., 2017. Prepubertal serum testosterone concentration in Vembur rams at different rearing system. The Indian J Vet SciBiotechnol. 12(4), 83-85. DOI:10.21887/ijvsbt.v12i4.7687

Selvaraju, S., Sivasubramani, T., Raghavendra, B.S., Raju, P., Rao, S.B., Dinesh kumar, D., Ravindra, J.P., 2012. Effect of dietary energy on seminal plasma insulin-like growth factor-I (IGF-I), serum IGF-I and testosterone levels,

semen quality and fertility in adult rams. Theriogenology. 78(3), 646–655. DOI: 10.1016/j. theriogenology.2012.03.010.

Smith, J.F., Parr, J., Murray, G.R,. Clarke, A.G., McDonald, R.M., Duganzich, D.M., 1998. Relationship between laboratory measures of ram sperm competence and field fertility. Proc New Zealand Soc Anim Prod.58, 181–185.

Verstegen, J., Iguer-Ouada, M., Onclin, K., 2002. Computer semen analyzer in andrology research and veterinary practice. Theriogenology. 57, 149-179.DOI: 10.1016/s0093-691x(01)00664-1

Walkden-Brown, S.W., Restall, B.J., Norton, B.W., Scaramuzzi, R.J., Martin, G.B., 1994. Effect of nutrition on seasonal patterns of LH, FSH and testosterone concentration, testicular mass, sebaceous gland volume and odour in Australian cashmere goats. J Reprod Fertil. 102, 351-360. DOI: 10.1530/jrf.0.1020351

World Health Organization (WHO), 1992. Laboratory manual for the examination of human semen and sperm- cervical mucus interaction. Cambridge: Cambridge University Press, Pp 11-13.

Zarazaga, L.A., Guzman, J.L., Dominguez, C., Perez, M.C., Prieto, R.,2009. Effects of season and feeding level on reproductive activity and semen quality in Payoya buck goats. Theriogenology. 71, 1316-1325.DOI: 10.1016/j.theriogenology.2009.01.007

Zarkawi,M.,1997. Monitoring the reproductive performance in Awassi ewes using progesterone radioimmunoassay. Small Rumin Res. 25, 291-294. https://doi.org/10.1016/S0921-4488(97)00011-4.

Zarkawi, M., Al-Daker, M.B., 2016. Body weight and reproductive parameters in fast and weak growing Awassi ram lambs during different age stages. Tropic Anim Health Prod. 48, 223-227. DOI: 10.1007/s11250-015-0929-x

Zarkawi, M., Al-Masri, M.R., Khalifa, K., 2003. An observation on yield and nutritive value of *Sesbania aculeata* and its feeding to Damascus does. Tropic Grasslands. 37, 187-192. Corpus ID: 55778391

Zarkawi, M., Al-Masri, M.R., Khalifa, K., 2005. Nutritive value of *Sesbania aculeata* grown on salty soil and its effect on reproductive parameters of Syrian Awassi ewes. Aust J Agric Res. 56,819-825.https://doi.org/10.1071/AR04301.