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Research Article

Reproduction Characteristics of Ouled Djellal Rams in a Semi-arid Area in Algeria

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ABSTRACT

The assessment of reproduction characteristics of an Algerian local sheep in a semi-arid zone was the objective of this study. Twenty rams of Ouled Djellal breed were used. The parameters analyzed were testosterone rate, testicular diameter, ejaculate volume, sperm concentration, mass motility, and percentage of live and dead spermatozoids (SPZ). The results obtained showed that the mean monthly testosterone rate was high throughout the year (4 ng/mL), increased in spring, peaked in summer (July) (4.9 ng/mL), and decreased in autumn and winter to reach a minimum value (January) of 1.9 ng/mL. Similarly, testicular diameter, ejaculate volume, sperm concentration and count showed high levels in spring and summer and decreased in autumn and winter. The maximum and minimum values were respectively, 7.09 cm, 1.55 mL, 4 billion/mL, 3.5, 79.37%, 21% and 5.46 cm, 0.80 mL, 2.50 billion SPZ/mL, 2.81, 77.36%, and 20.42%. Significant effects of season on serum testosterone levels, testicular conformation, ejaculate volume, and concentration were recorded in this study. Indeed, the rates of these parameters increased significantly ($P<0.05$) during spring/summer and decreased during autumn/winter except for motility, and the percentage of live and dead sperm. Testicular conformation was positively correlated with testosterone concentration, ejaculate volume, and semen concentration. To conclude, Ouled Djellal breed seemed not to be very sensitive to the photoperiod and little affected by high temperature.

Keywords: Sperm, Ram, Photoperiod, Local breed, Temperature, Season

Introduction

In Algeria, sheep breeding occupies by its numerical and socio-economic importance, a crucial place in breeding systems. It is concentrated in the steppe and constitutes an important animal resource for the country [1,2]. Mutton is the most popular consumed red meat especially in religious and traditional festivals. Eight main races have aired to date. Some of these breeds demonstrate strong adaptability to harsh environmental conditions. Among them, Ouled Djellal, also known as the white Arab race or Arbia, raised in arid and semi-arid regions and subject to a clear preference by farmers. Indeed, many studies reported the Ouled Djellal's good reproductive qualities and resistance to difficult conditions [3]. These particularly zootechnical performances contribute to the numerical productivity of the herds and therefore to obtaining good results in meat.

Improving the animal reproductive performances requires knowledge of the reproductive parameters of females and males. Ram sexual behavior can be influenced by many factors, including season, genetics, breed, hormonal condition, temperature, and nutrition. However, photoperiod is the main environmental factor affecting sheep reproduction [4]. Thus, the reproductive activity of most small ruminants, activated mainly by annual photoperiodism, exhibiting seasonal variations that extend proportionally with latitude. These variations are manifested in females by the existence of a period of seasonal anoestrus of variable duration depending on the breed and a favorable period for reproduction (sexual activity) [5]. While in the male, a period of low sexual activity could be manifested and is characterized by a decrease in the intensity of sexual behavior (libido), testicular size, hormonal secretion, and semen production both in quantity and quality [6]. The reproductive activity

Table 1. *Monthly averages of temperature, humidity, and photoperiod (daylight) during the study period*

Parameter	Month											
	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
Temperature (°C)	11	11.4	14.7	14.5	23	27.9	30.3	30.5	25.9	21.2	13.4	10.6
Duration of day (h)	9.75	10.83	11.93	12.5	13.93	14.5	14.33	13.6	12.5	11.7	10.2	9.83
Humidity (%)	79.3	67.1	66.7	71.5	51.5	49	40	41	52	62	72	78

of most animals native to temperate zones shows seasonal variations. These are more or less marked depending on the species and the amplitude of variation of the photoperiod. Small ruminants show periods of non-activity in the long photoperiod and periods of activity in the short photoperiod [7]. In sheep/goats, so-called "short-day" species, the photoperiod is certainly the main cause of the seasonal variations in reproduction observed for all breeds in Northern Europe. However, in tropical and subtropical regions, variables related to temperature and nutrition also appear to affect reproductive physiology in animals [5].

In rams, during reproductive life, body weight, scrotal circumference, testosterone levels, and sperm production change under the influence of several internal and external factors. Therefore, ram reproduction data showed a complex relationship between the development of the neuroendocrine system and environmental conditions [8]. Seasonal variation in mammalian reproduction results from adaptation to annual environmental changes [9]; thus, they are an important factor influencing sperm quantity and quality. This is why the knowledge of these reproductive factors makes it possible to genetically improve the breed and increase the herd productivity [10]. The present study aimed to evaluate the influence of the season on testosterone concentration, testicular diameter, and sperm production in Ouled Djellal rams in Algerian semi-arid zone. Knowledge of this information is important insofar as it should determine the favorable or inferred periods for the spermogram, the choice and use of parents during the year and thus improve the success of insemination. This work also leads to the promotion of artificial insemination using seeds from local breeds.

Materials and Methods

Ethical Statement

All the animal studies were conducted with the utmost regard for animal welfare, and all animal rights issues were appropriately observed. No animal suffered during the course of the work. All the experiments were carried out according to the guidelines of the Institutional Animal Care Committee of the Algerian Higher Education and Scientific Research (Agreement Number 45/DGLPAG/DVA.SDA.14).

Study Area

The study was conducted at a private sheep farm located 20 km from the city of Chlef (Algeria). It is an agricultural farm, combining sheep breeding and crops (cereals and fodder). Sheep population is large and unique, around 1100 heads (350 ewes, 750 heads between rams and lambs). The entire herd is essentially of Ouled Djellal breed. The farm has 4 breeding buildings housing the different categories of animals. The area is located between 36°10′ North latitude and 1°20′ East longitude and at an altitude of 86 m. Chlef region, located in the North of Algeria (200 kilometers from the capital Algiers), is known for its semi-arid Mediterranean climate with mild little rainy winters and hot, dry summers. *Table 1* represents the average monthly variations in temperature, humidity, and photoperiod of the region during the study year (2021).

Experimental Herd

In this current work, 20 pubescent Ouled Djellal rams born on the farm and raised in the region were used. The animals selected all responded to the standard type of the Ouled Djellal breed (physical characteristics and measurements). It is an entirely white breed, with fine wool and tail, high waist and long legs, suitable for walking (*Fig. 1*). The ears are long and horizontal. The horns, present in both sexes in general, may be missing in ewes. The ram weighs up to 80 kg and the ewe 60 kg [11]. This sheep is large : height at the withers, body length, and chest circumference are respectively, 80 cm, 84 cm, and 40 cm for the male and 74 cm, 67 cm, and 35 cm for the female. The rams in our study were 3 to 4 years old, with a weight varying between 55 and 60 kg (56.8±2.1 kg). The animals

Fig 1. Ouled Djellal breed on the farm

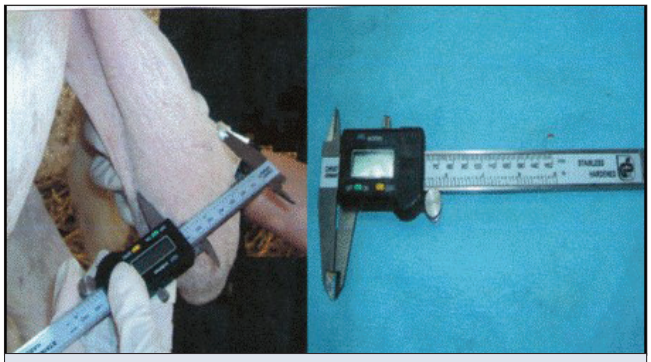

Fig 2. Testiclar anteroposterior diameter measurement

were kept in a free stall, subject to seasonal variations and environmental factors in the study region. They received a constant diet, consisting of *ad libitum* oat vetch hay and approximately 600 g of barley concentrate per head per day, and benefited from ad libitum watering. The rams were isolated from the ewes in open barns and kept out of breeding for one year. All animals were previously treated against diseases and classic parasites of sheep and were free of any pathology including brucellosis.

Blood Samples and Testosterone Assay

Blood samples were taken weekly from the jugular vein of each ram for one year (from January to December). The blood was immediately centrifuged and the serum obtained was immediately frozen at -20°C until testosterone assay. Serum testosterone concentration was obtained using the ELISA technique, as previously described [12].

Testicular Diameter Measurement

Every week and for a year, the testicular anteroposterior diameter was measured using a caliper [6,13]. These measurements were taken for each ram and always by the same operator *(Fig. 2)*.

Collection and Analysis of Semen from Rams

Semen collection was performed using an artificial vagina (made and suitable for sheep) in the presence of female in heat and attached to a special support. The rams selected were dressed for artificial vagina collection for a period of 2 to 3 months. The collections lasted a year, at a rate of one collection per week and per ram, carried out early in the morning. The characteristics of the sperm recorded were: Volume (recorded directly in the graduated glass collection tube); Concentration (spermatozoa/mL of ejaculate), measured by a spectrophotometer after 1/400 dilution in an isotonic sodium chloride solution [14]. Sperm mass motility was obtained by examining a drop of semen under a microscope equipped with a hot plate at 37°C. To evaluate the intensity of the waves formed by the movements of the spermatozoa, a score was thus assigned ranging from 0 to 5 [15]. The vitality of the spermatozoa was provided by counting after staining with Eosin-Nigrosin, to determine the percentage of living and dead spermatozoa (Eosin negative). The reading was done under a microscope. Live sperm stain white and dead stain pink or red [16].

Statistical Analysis

Results were collected monthly and presented as mean and standard deviation. ANOVA was used to determine the seasonal variations of the parameters, the correlations, and the PCA (Principal Component Analysis) to explain the variance of these factors. Pearson's significance test (P) was performed to check for variations. The differences detected were considered as highly significant when $P<0.0001$ and significant when $P<0.05$. XLSTAT software version 2002 was used for all these analyses.

Results

Seasonal Variations in Serum Testosterone

The mean baseline testosterone level of Ouled Djellal rams in this study was 3.34±0.55 ng/mL. However, over the twelve months of analysis, the concentrations were characterized by very marked monthly variations *(Table 2)*.

Indeed, a rise in concentration from February (2.98±0.26 ng/mL), to reach a peak (maximum value) in July (4.93±0.39 ng/mL) was noticed. A highly significant drop of approximately 45% was revealed in October (2.65±0.38 ng/mL), which stabilized in November-December and decreased again to reach its minimum value in January (1.92±0.29 ng/mL).

Variations in Antero-posterior Testicular Diameter

The mean value of the testicular diameter in this work was 6.23±018 cm. The previous table reported the rates

Table 2. *Monthly means of serum testosterone and testicular diameter of the 20 Ouled Djellal rams*

Parameter	Month											
	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
Testosterone (ng/mL)	1.92±0.29	2.98±0.26	3.57±0.51	3.7±0.62	3.80±0.56	4.38±0.72	4.93±0.39	3.75±0.81	3.45±0.73	2.65±0.38	2.55±0.45	2.50±0.82
Testicular diameter (cm)	5.46±0.19	6.32±0.15	6.49±0.21	6.58±0.13	6.62±0.17	6.98±0.14	7.09±0.12	6.80±0.17	5.78±0.21	5.60±0.22	5.56±0.19	5.49±0.20

Table 3. *Results of ANOVA analysis*

Season	Test. (ng/mL)	TD (cm)	VOL (mL)	SC (10 ^{9}/mL)	MM	LS (%)	DS (%)
Spring	4.025b	6.7825b	1.4175a	3.9775a	3.1375a	80.95a	20.4275ab
Summer	4.7375a	7 187a	1.505a	3.8275a	3.0875a	75.45b	21.155a
Autumn	2.67c	5.39c	0.980b	2.9375b	2.6725b	77.02b	20.3975b
Winter	2.3225c	5.46c	0.9075b	2.825b	2.69b	75.46b	20.04b
R^{2-}	0.9315	0.9377	0.9285	0.9586	0.6219	0.7807	0.5110
F	54.4014	60.1677	51.9621	92.6606	6.5801	14.2370	4.1808
Pr > F	<0.0001	<0.0001	<0.0001	<0.0001	0.0070	0.0003	0.0305
Significance	HS	HS	HS	HS	VS	HS	S

Test.: Serum testosterone, TD: testicular diameter, VOL.: ejaculate volume, SC: Sperm concentration, MM: Mass Motility, LS: live Sperm, DS: Dead Sperm, HS: highly significant, VS: very significant, S: significant, R^2: coefficient of determination, F: ratio of two standard deviations, Pr: probability

of the seasonal variations in this diameter measured in Ouled Djellal rams. The variations were similar to those of blood testosterone. The seasonal influence on testicular volume and testosterone was statistically highly significant (P<0.0001) *(Table 3)*. The lowest averages were recorded in autumn/winter (September to January) and the highest in spring/summer, peaking in July.

The analysis of the monthly variations of testosterone and testicular diameter with those of the climatic factors showed a close parallelism between the profiles of seasonal evolutions of these parameters and the length of the day. It was minimum in autumn/winter then increased at the same time as the photoperiod to be maximum in spring/summer.

Seasonal Variations in Sperm Characteristics

All the rams could be sampled for semen. Thus, 45 to 48 samples were taken from each ram in this study. Semen from Ouled Djellal rams presented a milky white

Table 4. *Individual mean values of the sperm parameters*

Ram	Age (year)	LW (kg)	V (mL)	SC (x10^9/mL)	MM	LS (%)	DS (%)
1	3	56.0±0.8	1.10±0.60	2.99±0.15	2.77±0.17	79.20±10.20	27.00±5.50
2	3	56.7±0.6	1.40±0.35	3.28±0.32	4.10±0.23	80.00±15.00	17.20±4.90
3	3	60.2±0.9	1.38±0.32	3.22±0.39	3.79±0.15	89.00±9.10	9.10±5.21
4	4	55.0±1.0	1.45±0.21	2.60±0.18	3.85±0.10	76.21±12.23	28.45±3.95
5	4	57.7±0.7	1.25±0.75	4.00±0.30	2.89±0.09	59.50±10.00	45.21±3.00
6	3	58.1±1.0	1.27±0.25	2.70±0.50	3.00±0.16	56.40±19.50	30.23±6.50
7	3	59.2±0.5	1.32±0.66	3.00±0.40	3.10±0.10	90.21±15.65	8.80±7.10
8	4	60.5±2.1	1.30±0.55	3.25±0.28	3.10±0.11	88.22±12.00	13.00±6.32
9	4	55.1±1.7	1.39±0.35	3.00±0.45	2.98±0.20	70.00±17.21	23.21±6.98
10	3	55.0±0.6	1.55±0.10	3.05±0.35	3.21±0.15	73.45±18.54	22.25±4.23
11	4	56.2±0.3	1.10±0.12	2.99±0.32	3.00±0.25	90.00±8.88	8.12±4.21
12	3	58.0±0.6	1.20±0.15	3.87±0.20	3.00±0.16	92.00±7.00	8.00±3.98
13	4	59.0±0.7	1.28±0.55	4.20±0.25	4.20±0.13	68.22±22.00	25.00±2.99
14	4	55.5±0.2	1.55±0.45	4.00±0.45	3.90±0.14	49.50±12.00	40.00±3.99
15	3	55.4±0.6	1.55±0.21	3.20±0.30	3.54±0.21	87.96±12.00	26.98±6.32
16	3	57.1±0.4	1.36±0.32	3.78±0.20	2.79±0.13	89.00±10.87	15.00±5.52
17	4	57.0±0.8	1.30±0.24	3.90±0.40	4.20±0.14	91.21±9.00	8.88±5.21
18	3	59.2±1.0	1.40±0.36	2.98±0.27	2.97±0.25	79.65±8.96	20.12±4.51
19	4	60.0±0.9	1.08±0.60	3.60±0.36	2.79±0.10	85.56±12.12	12.00±5.00
20	3	60.0±0.3	1.10±0.50	3.25±0.54	3.21±0.21	80.32±10.56	16.54±3.32
Avg.	3.8±0.7	57.5±1.7	1.40±0.43	3.73±0.30	3.31±0.15	78.78±12.04	20.25±4.93

LW: Live weight, V: ejaculate volume, SC: sperm concentration, MM: mass motility, LS: live spermatozoa, DS: dead spermatozoa

Table 5. *Monthly means of the sperm values in rams*

Parameter	Month											
	Jan.	**Feb.**	**Mar.**	**Apr.**	**May**	**Jun.**	**Jul.**	**Aug.**	**Sep.**	**Oct.**	**Nov.**	**Dec.**
Volume of ejaculate (mL)	0.80±0.31	1.15±0.20	1.30±0.44	1.43±0.50	1.51±0.27	1.55±0.43	1.53±0.29	1.45±0.35	1.12±0.30	1.05±0.26	1.00±0.15	0.90±0.60
Sperm concentration ($X10^9$/mL)	2.50±0.52	3.19±0.73	3.66±0.52	3.69±0.58	3.75±0.42	3.90±0.30	4.00 ± 0.33	3.12±0.27	3.02±0.29	2.98±0.26	2.93±0.15	2.84±0.29
Mass motility	2.73±0.10	3.10±0.21	3.37±0.32	3.60±0.27	3.74±0.11	3.75±0.15	3.59±0.25	3.00±0.18	2.75±0.19	2.76±0.11	2.78±0.13	2.75±0.22
Live sperm (%)	74.00±10.05	76.20±9.25	80.90±10.02	82.23±8.77	85.00±12.21	82.00±10.00	75.10±8.95	71.00±9.90	79.50±11.10	80.10±14.05	79.40±13.20	75.00±12.00
Dead sperm (%)	20.30±5.30	20.23±4.12	19.95±6.01	20.15±7.10	20.00±6.00	21.01±4.50	22.00±3.70	22.90±3.30	21.15±6.50	20.62±3.98	20.10±4.00	20.15±6.14

Table 6. *Pearson correlation matrix between variables*

Variables	**Test.**	**TD**	**V**	**SC**	**MM**	**LS**	**DS**
Test.	**1**	0.9850	0.9897	0.9338	0.9304	0.2271	0.8790
TD	0.9850	**1**	0.9900	0.9541	0.9655	0.2653	0.8005
V	0.9897	0.9900	**1**	0.9753	0.9720	0.3529	0.8039
SC	0.9338	0.9541	0.9753	**1**	0.9942	0.5408	0.6595
MM	0.9304	0.9655	0.9720	0.9942	**1**	0.4983	0.6433
LS	0.2271	0.2653	0.3529	0.5408	0.4983	**1**	-0.1648
DS	0.8790	0.8005	0.8039	0.6595	0.6433	-0.1648	**1**

Test.: serum testosterone, DT: testicular diameter, V: ejaculate volume, SC: sperm concentration, LS: live spermatozoa, DS: dead spermatozoa

appearance in 70% of the cases, against 30% having a creamy appearance.

The mean ejaculate volume of rams was 1.40±0.43 mL (*Table 4*). It is varied between 1.10 mL (minimum value) to 1.55 mL (maximum value). This parameter also underwent highly significant seasonal variations (P<0.0001) *(Table 3)*. The volume of ejaculate was greater in spring and summer than in autumn and winter, respectively (1.4±0.4 mL, 1.5±0.4 mL versus 1.1±0.2 mL, 1.0±0.4 mL) *(Table 5)*.

The average concentration in this test was found to be 3.73±0.30 billion sperm per milliliter of semen, with respective minimum and maximum values of 2.60 and 4.1 billion sperm per milliliter of ejaculate. According to the data by month, the concentration increased from March to reach the seasonal peak in June (4.0±0.3 billion) and decreased from July reaching a minimum value in January (2.5±0.5 billion). Seasonal variations of this parameter were also highly significant (P<0.0001). It was higher when the photoperiod was increasing (spring/summer) than when it was decreasing (autumn/winter). Semen from Ouled Djellal rams was more concentrated in spring and summer than in autumn and winter.

The mean values of mass motility, percentage of live and dead spermatozoa obtained in this study were 3.31±0.15, 78.78±12.04 and 20.25±4.93, respectively *(Table 4)*.

Table 3 showed that mass motility was very significantly affected by the season (P<0.001). In fact, the highest values were recorded in spring/summer (3.58±0.23, 3.44±0.19) and the lowest in autumn/winter (2.76±0.14, 2.89±0.18).

Also, the analysis of the average monthly results in *Table 5* demonstrated a highly significant influence of the season (P<0.0001) on the percentage of live spermatozoa. The maximum value (85.71±12.21%) was observed during spring, mating period in the region and the minimum value in winter (74±10.05%). The seasonal mean results of the percentage of dead spermatozoa showed a slight variation from month to month *(Table 5)*. Indeed, the minimum extreme values in March (19.95±6.01%) and maximum in August (22.90±3.30) were revealed over the study period and did not show a very significant difference (P<0.05).

Finally, the results obtained showed that the rates of all the improved parameters evolved with the photoperiod and the temperature. Their monthly means increased from February, reaching a maximum in June (4.93±0.34 ng/mL, 7±0.12 cm, 1.55±0.43 mL, 4±0.30x10 9 Spzs/mL, 3.75±0.15, 85±10%), then decreased from September to reach their minimums in January (1.92±0.29 ng/mL, 5.46±0.19 cm, 0.8±0.31 mL, 2.50±0.52x10^9 Spzs/mL, 2.73±0.10, 73±10.5%). So, it appears that the Ouled Djellal ram produces large quantities of sperm all year round. It

is more important, more concentrated, more mobile and vital from February/March (increasing photoperiod).

Correlations

According to the PCA examination, more than 90% of the variations of the parameters were explained by the months of the year. Indeed, according to *Table 6*, the spring and summer seasons were positively correlated (r=0.80) with concentration, motility, ejaculate volume, percentage of live spermatozoa and testicular diameter. Also, the analysis of the correlation matrix of *table 3*, showed a highly significant difference (P<0.0001) between the season and the testicular diameter, the volume, the concentration, and the % of live spermatozoa and a very significant difference (P<0.001) between the season and the mass motility, while the difference between the season and the % of spermatozoa was a bit significant (P<0.05).

The relationship between the testicular measurements and the concentration of testosterone made by the correlation coefficients are presented in *Table 6*. The testicular diameter had a very high and positive correlation with the testosterone rates (r=0.98). The latter had high and positive correlations ranging from 0.93 for mass motility and sperm concentration and 0.98 for ejaculate volume. Similarly, testicular diameter showed very high and positive correlations between all sperm characteristics, with volume (r=0.99), sperm concentration (0.95), and mass motility (r=0.96).

Discussion

In this study, serum testosterone underwent significant seasonal variation. It was higher from March to July than from October to January. This pattern of variation is similar to those described by Darbeida et al.[17] and Belkadi et al.[14] in the same breed in Algeria and by Issa et al.[18] in the Fulani and Tuareg breed in the Sahelian climate.

According to our data, the testicular diameter measured in the Ouled Djellal ram was always high during the year. According to several authors, it depends on age, body weight, diet, and sexual season [12]. This result could be explained by the fact that the rams of our sample were all adults, of good body conformation and raised in good food and sanitary conditions. This parameter reflects sperm production and is a good indicator of male fertility; its heritability improves reproductive parameters [19]. Indeed, the scrotal circumference is an indicator of the amount of testicular spermatogenetic tissue, which reveals the maximum potential for sperm production. It is possible that larger testes contain more androgen-producing tissue (Leydig cells), where high levels of stimulation promote the growth and development of spermatogenic tissue [20]. These authors' conclusions were confirmed in this study by the strong positive correlations obtained between testicular diameter and sperm characteristics. Also, Belkhiri et al.[21], reported that measurement of testicular diameter in rams could be used by Breeding Centers to select suitable males for artificial breeding. Thus, any factor that influences testicular volume affects the reproductive efficiency of rams. The testicular conformation was highly affected by the season. Sexual activity peaked in spring/summer despite the increase in day length. Unlike sheep breeds in temperate zones where sexual activity increases in autumn when the photoperiod decreases. These results are in agreement with those of Kafi et al.[6] and Tabbaa et al.[22] in the Awassi breed, of Milczewski et al.[23] in the Suffolk breed, and of El Bouyahiaoui et al.[24] in the Tazegzawt ram.

Choosing an effective semen collection method is the first step in establishing a sperm cryopreservation bank. The artificial vagina collection method was used in this study because it generally results in better quality semen. The milky and creamy appearance of the semen samples collected from the Ouled Djellal rams in our work showed that the semen from this breed was mostly normal, similar to that of the majority of sheep breeds [18].

The Ouled Djellal rams produced a sperm volume included in the range of averages of 1 to 1.5 mL given for sheep [25]. In another study performed on South African indigenous rams, Ngcobo et al.[12] reported no significant difference in semen volume during the spring (0.88 mL), summer (0.91 mL), autumn (0.92 mL), and winter (0.92 mL) seasons. This may be due to the fact that Ouled Djellal rams are of significant size and testicular conformation, because the average volume of ejaculate increased according to body mass and testicular conformation. Indeed, in this study, this last parameter was found to be very strongly correlated with the volume of ejaculate [12,21].

The mean value of the spermatozoa concentration obtained is within the range of rates reported in sheep, which is between $2x10^9$-$6x10^9$ spermatozoa/mL [26,27]. This mean value is significantly higher than those of D'man [28], Noire de Thibar rams from Tunisia [29], and Djallonké from Ivory Coast [27], which were respectively $1.8x10^9$ Spz/mL, 2.9 $0x10^9$ Spz/mL, $3x10^9$ Spz/mL, and $2.55x10^9$ Spz/mL. This value is close to the concentration of semen in white Fulani and Tuareg rams, which were respectively $3.7\pm1.3x10^9$ Spz/mL, and $3.6\pm0.4x10^9$ Spz/mL [30]. This sperm production is mainly due to the high testicular weight of the animals in general, a consequence of the large format of the Ouled Djellal ram. This can also be explained by the very strong positive correlation obtained in this current work between testicular diameter and sperm production. Indeed, it is well established that the production of spermatozoa per gram of testis is a characteristic of the species and of the breed.

The average value of the mass motility recorded in this study is considered good in comparison with the value 4, from which this parameter is evaluated as acceptable [15] and the sperm in this case can be used for artificial insemination. The rate obtained in this study is close to that of European rams such as Romanov and Ile-de-France (3.5 to 4.1) [31]. However, it is higher than those reported for white Fulani and Tuareg rams from the Sahel, which were respectively 2.89±1.2 and 2.90±0.1 [30]. The determination of this parameter constitutes, in the current state of knowledge, a criterion for sorting ejaculations to be used for insemination and breeding animals [15]. Fluctuations in mass motility were not really affected by photoperiod and temperature as already shown by Colas [31] and Aller et al.[16]. This is in agreement with the finding of Ghozlane et al.[32] and Kafi et al.[6], who demonstrated in a study of Ouled Djellal rams in Algeria and Persian Karakul rams in Iran, that photoperiods and high temperatures did not influence mass motility and concluded that sperm quality was higher during and towards the end of summer.

The average monthly results of the percentage of live spermatozoa showed a significant difference between those observed during the spring/summer and autumn/winter seasons. Although the maximum average was recorded in spring/summer, the season corresponding to the breeding period in this region, the viability of the ram's spermatozoa was maintained during the year, since the average values were always above 70%. Benia et al.[33] did not note a decrease in sperm viability in Rembi breed rams (raised under arid zone conditions in Algeria) during long and short photoperiod periods. The viability of the spermatozoa of these rams, like that of the Ouled Djellal rams in our sample, was maintained throughout the year with peaks in spring/summer.

The overall average of dead spermatozoa found in the present study is within the standards accepted for good quality sperm (between 20 and 25%) [31]. This value was consistent with the findings of Aller et al.[16] who claimed that good quality sperm should not contain more than (25%) of dead sperm. If the rate exceeds 25% of the total number in an ejaculate, a reduction in fertility can be anticipated.

The values of all the sperm characteristics obtained in this work can confirm that the semen of the Ouled Djellal rams collected under the environmental conditions of Chlef region is of good quality. Indeed, the results obtained with a collection rate of once a week showed that the sperm production potential of this breed is the best compared to that of other breeds produced at the same rate. This strong production potential allows the intensification of the productivity of the sheep herds insofar as the rams will be used at a faster rate. In view of these seasonal variations in the year of study, this period (spring/summer) may seem favorable to the spermogram of the Ouled Djellal ram and it corresponds to the period of traditional struggle of sheep herds in the region. The profile of variation of all the parameters is explained as well as the strong positive correlations obtained between them, confirms the imbrication in the ram of the endocrine (hormone production) and exocrine activity of the testicle (spermatogenesis).

In this study, it was not possible to observe the effect of the age and the weight of the rams on these parameters because the animals were chosen as all adults and had on average the same age (3 to 4 years) and the same weight (55 to 60 kg).

Also, according to our data, the period from February to July appears to be favorable to the spermogram of the Ouled Djellal ram even if seasonal effects have not been observed. This variation profile has also been detected in the Djallonké ram from Ivory Coast [27], in the same breed in Algiers region [14,32], and in several rams from tropical climates [34]. Thus, the ram of the Ouled Djellal breed showed a particular behavior: a slight decrease in its sexual activity in autumn/winter, despite a theoretically favorable photoperiod and an increase in spring/summer. This indicates that contrary to what is accepted, the reproductive abilities of Ouled Djellal rams in Chlef region (latitude 36°N) are not subject to photoperiod variations. The qualities of the sperm of Ouled Djellal rams were not really degraded during the year, because the bad rates of spermogram parameters were not obtained. The registered sperm therefore retained, with regard to all the measured parameters, good fertility throughout the year. This corroborates the observation made in Ouled Djellal ewes who lamb all year round [35]. This feature highlighted in this study is different from that of most other breeds of rams living in Europe and North America. In these regions, the peaks of sexual activity occur from September to November and births at the end of winter and spring [36,37].

This low seasonality is also observed in the fat-tailed Awassi breed in Turkiye (Lat. 36 to 42°N) [38], the Romney breed in New Zealand (Lat. 40°N) [39] and the Southdown breeds, and Targhee in Ohio in the United States (Lat. 40°N) [40].

Presumably, the seasonal variations in the activity of the Ouled Djellal ram testis appear, result from parallel variations in the production of hypothalamic-pituitary-gonadal hormones and from a modification of the receptivity of the interstitial gland to pituitary stimuli. In the ram, Beltrán-Frutos et al.[41], distinguished in the annual testicular cycle, three main phases called "regression" (minimum testicular weight), "development" and "activity" (maximum testicular size) and showed that

the LH-RH peaks are fewer during the "regression" phase. Therefore, the intensity of sexual activity manifested in spring/summer in the Ouled Djellal ram can be explained by the increase in the total quantity of LH-RH released by the hypothalamus during the testicular "development" phase (in spring/summer). Also, in the Ile de France ram, very high correlations are presented between the pituitary LH content and the weights of the testicle and the accessory glands. These parameters show lower values from the end of November to the end of January (in autumn and winter) [42].

These observations made believe of that the seasonal variations of the parameters, manifested in the Ouled Djellal ram, are secondary to parallel modifications of the activity of the hypothalamic-pituitary-gonadal system. The increase in the values of the parameters taken into account in spring/summer is due to the spring reactivation of the testis (development and activity phase) caused by an increase in LH-RH and LH secretions. Also, to the possibility of seasonal variations in the sensitivity of the pituitary to LH-RH. Indeed, frequent photoperiodic manipulations have proven that the receptivity of the pituitary to the hypothalamic factor is greater in the testicular activity phase (spring/summer) than in the regression phase (autumn/winter), leading to much greater LH-RH discharges in the activity phase and the endogenous release of LH by frequent episodic peaks [12]. Therefore, it appears that the frequency of LH-RH discharges is the factor determining the cycle of variations in spermatogenesis in rams [43].

Seasonal variations in prolactin levels have also been described in Ouled Djellal rams [17], with high levels in spring/summer and much lower levels in winter. Considering the stimulating role of prolactin on the sexual sphere and in particular on the testicular function, its intervention in the determinism of the seasonal variations of the sexual activity of the ram must be granted.

Many studies have highlighted the importance of the quality and quantity of food on sexual activity. The restriction or insufficiency of the food ration produces a decrease in spermatogenesis and in testosterone production in rams [44]. Since this factor is considered to be determining in the sexual activity, in our study and because animals received throughout the duration of the experiment, a sufficient and constant diet both in quantity and in quality, the results have revealed no variation nor influence.

In this study, there is also a parallel evolution of the reproductive activity of the ram and the average monthly temperatures. However, many authors have reported that in mid-latitudes (case of the Chlef region), the thermal environment is not the main factor influencing sexual activity, and temperature fluctuations do not alter the reproductive pattern of sheep [45]. Rather, temperature effects are in general related to an increase in the animals' body temperature. But Ouled Djellal breed seems little affected by high summer temperatures and is perfectly adapted to the thermal environment of its biotope. In the other hand, other studies have shown that exposure of males to extreme temperatures can constitute heat stress and can negatively affect their fecundity [46]. This may partly explain the slight decrease in sperm parameters in the Ouled Djellal ram during the months of July and August.

At the end, Ouled Djellal rams present a little seasonal infulence on reproduction, similar to that of the ewe of the same breed. Sexual activity is higher in spring/summer and lower in autumn/winter. This finding is not in agreement with observations generally made in sheep, which known as sexually short-day species and in which photoperiod plays an essential role in the regulation of seasonal reproductive activity. This breed therefore seems not to be very sensitive to the photoperiod and little affected by high temperatures. The sperm qualities throughout the year, are compatible with normal fecundity. These results should be considered as an incentive to use Ouled Djellal rams for year-round breeding.

Availability of Data and Materials

The datasets during and/or analyzed during the current study available from the corresponding author (N. Mimoune) on reasonable request.

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Ethical Statement

All the experiments were carried out according to the guidelines of the Institutional Animal Care Committee of the Algerian Higher Education and Scientific Research (Agreement Number 45/ DGLPAG/DVA.SDA.14).

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Author Contribution

MT, NAI, NM: Conceptualization, Methodology, Validation, Formal Analysis, Investigation, Writing - Original Draft, Writing - Review & Editing. DK: Supervision, Investigation.

References

1. Ait Issad N, Abdelouahed K, Mimoune N, Bekhouche S, Boubeuker R, Hamoudi Adjmi H, Ait Hamouda TG, Degui D, Kaidi R, Khelef D: Molecular detection of *Toxoplasma gondii* in ewes placenta in Northeastern Algeria. *Kafkas Univ Vet Fak Derg*, 28 (2): 267-274, 2022. DOI: 10.9775/kvfd.2021.26887

2. Rahmani, M, M, Mimoune N, Saidi R, Benayada A, Mettache Z, Saci, N, Kaidi R: Study of helminths in sheep in the Laghouat region in Southern

Algeria. *Vet Stanica,* 54 (1): 59-67, 2023. DOI: 10.46419/vs.54.1.6

3. Dekhili M: Phenotypic and genetic parameters of Ouled-Djellal sheep reproduction (Algeria). *Arch Zootech,* 63 (242): 269-275, 2014. DOI: 10.4321/S0004-05922014000200005

4. Ramírez Ramírez AI, Delgado Tiburcio GI, Cruz-Espinoza F, Herrera-Corredor AC, Gallegos-Sánchez J: Photoperiod and its relationship to sheep reproduction. *Agro Productividad,* 2021. DOI: 10.32854/agrop.v14i10.1620

5. Starič J, Farci F, Luridiana S, Mura MC, L, Pulinas G, Cosso V, Carcangiu V: Reproductive performance in three Slovenian sheep breeds with different alleles for the *MTNR1A* gene. *Anim Reprod Sci,* 216, 106352, 2020. DOI: 10.1016/j.anireprosci.2020.106352

6. Kafi M, Safdarian A, Hashemi M: Seasonal variation in semen characteristics, scrotal circumference and libido of Persian Karakul rams. *Small Ruminant Res*, 53 (1-2): 133-139, 2004. DOI: 10.1016/j.smallrumres.2003.07.007

7. Arjoune A, Alsaleh AB, Messaoudi SA, Chelbi H, Jelassi R, Assidi M, Najar T, Haddad B, Sirard MA: Analysis of *MTNR1A* genetic polymorphisms and their association with the reproductive performance parameters in two Mediterranean sheep breeds. *Animals*, 13 (3):448, 2023. DOI: 10.3390/ani13030448

8. Maksimovic N, Hristov S, Stankovic B, Petrovic, MP, Mekic C, Ruzic-Muslic D, Cardo-Petrovic M: Investigation of serum testosterone level, scrotal circumference, body mass, semen characteristics and their correlations in the development of MIS lambs. *Turk J Vet Anim Sci,* 40, 53-59, 2016.

9. Corte Pause F, Crociati M, Urli S, Monaci M, Degano L, Stradaioli G: Environmental factors affecting the reproductive efficiency of Italian Simmental young bulls. *Animals (Basel)*, 12 (18):2476, 2022. DOI: 10.3390/ani12182476

10. Seifi-Jamadi A, Zhandi M, Kohram H, Luceño NL, Leemans B, Henrotte E, Latour C, Demeyere K, Meyer E, Van Soom A: Influence of seasonal differences on semen quality and subsequent embryo development of Belgian Blue bulls. *Theriogenology,*158, 8-17, 2020. DOI: 10.1016/j.theriogenology.2020.08.037

11. Kerboua M, Feliachi K, Abdelfettah M, Ouakli K, Selhab F, Boudjakdji A: National report on animal genetic resources: Algeria. 1-46, Ministry of Agriculture and Rural Development, National Commission An GR 2003.

12. Ngcobo JN, Nedambale TL, Mpofu TJ, Nephawe KA, Chokoe TC, Ramukhithi FV: Seasonal variations in semen quality, testosterone levels, and scrotal size following dietary flaxseed oil and ascorbic acid in South African indigenous rams. *Animals*, 13 (7):1213, 2023. DOI: 10.3390/ani13071213

13. Haro M, Zongo M, Bayala B: Morpho-biometric characterization of the testicle of the Sahelian goat. *Int J Biolog Chem Sci*, 16 (2): 713-720, 2022. DOI: 10.4314/ijbcs.v16i2.16

14. Belkadi S, Safsaf B, Heleili N, Tlidjane M, Belkacem L, Oucheriah Y: Seasonal influence on sperm parameters, scrotal measurements and serum testosterone in Ouled Djellal rams in Algeria. *Vet World*, 10 (10): 1486-1492, 2017. DOI: 10.14202/vetworld.2017.1486-1492

15. Baril G, Chemineau P, Cognié Y, Guerin Y, Leboeuf B, Orgeur P, Valet JC: Training Manual for Artificial Insemination in Sheep and Goats. FAO Study Prod. Health Anime, 83, 231, 1993.

16. Aller JF, Aguilar D, Vera T, Almeida GP, Alberio RH: Seasonal variation in sexual behavior, plasma testosterone and sperm characteristics of Argentine Pampinta and Corriedale rams. *Scope J Agric,* 10 (2): 345-3525, 2012. DOI: 10.5424/sjar/2012102-389-11

17. Darbeida H, Brudieux R, Ravault J: Annual variations in plasma concentrations of prolactin and testosterone in Bélier Ouled-Djellal, in Algeria. *Sci Life,* 299 (19):789, 1984.

18. Issa M, Yenikoye A, Marichatou H, Banoin M: Spermogram of two-tone Fulani and Tuareg rams: Influence of genetic type and season. *Rev Elev Med Vet,* 54 (3-4): 2001. DOI: 10.19182/remvt.9784

19. Ramírez-López C, Rugeles-Pinto C, Guimaraes JD, Vergara-Garay O: Relacion entre biometríatestu lar y circunferenciaescrotal en toretes de la raza Nelore en Brasil. *Revista Científica*, 26 (1): 49-54, 2016.

20. Preston BT, Stevenson IR, Lincoln GA, Monfort SL, Pilkington JG and Wilson K: Testicular size, testosterone production and reproductive behavior in a natural mammalian mating system. *J Anim Ecol,* 10, 1365-2656, 2012. DOI: 10.1111/j.1365-2656.2011.01907.x

21. Belkhiri Y, Bouzebda-Afri F, Bouzebda Z, Mouffok C: Effects of age and season on sexual parameters in adult rams used in an artificial insemination center. *Glo Vet,* 18 (1):3140, 2017.

22. Tabbaa MJ, Kridli RT, Al-Ghalban A, Barakeh FS: Age-related changes in scrotal circumference and some sperm characteristics in Awassi rams. *Anim Reprod,* 3 (4): 431-438, 2006.

23. Milczewski V, Chahad-Ehlers S, Spercoski KM, Morais RN, Soccol VT: Quantification of the effect of seasonality on testicular function in low-latitude Suffolk rams. *Small Ruminant Res,* 124, 68-75, 2015. DOI: 10.1016/j.smallrumres.2014.12.012

24. El Bouyahiaoui R, Oulkhier F, Fantazi K, Bouraada A, Cherragui N, Sebbagh L, Herkat S: Study of seasonal variation in testicular morphobiometry and sexual behavior in the Tazegzawt ram. *Search Agro INRA*, 20 (1): 61-75, 2022.

25. Dotche IO, Kiki P, Govoeyi B, Dahouda M, Antoine-Moussiaux N, Dehoux JP, Mensah GA, Farougou S, Thilmant P, Karim IYA, Koutinhouin B: State of play on animal artificial insemination in West African countries. *J App Biosc*, 143, 14712-14730, 2019.

26. Ghorbankhani F, Souri M, Moeinin, MM, Mirmahmoudi R: Effect of nutritional status on sperm characteristics, testes size and serum tester concentration in Sinjabi ram lambs during natural breeding season. *Anim Reprod Sci*, 153, 22-28, 2015. DOI: 10.1016/j.anireprosci.2014.12.006

27. Lavry GN, Coulibaly M'B, Offoumou AM, Datte JY: Three-year monthly variations in the spermogram of Djallonké rams in the forest region of Côte d'Ivoire. *Rev Elev Med Vet Country Too*, 69 (3): 111-116, 2017.

28. Chafri N, Mahouechi M: Effect of food level before and after weaning on sperm production in D'man rams. *Meeting Search Rum,* 16:318, 2009.

29. Hammami M, Hammami M, Rouissi H, Werghi H. and Tarkhani I: Diagnosis and analysis of the situation of black sheep breeding in Thibar in northern Tunisia. *Live Res Ru Develop,* 26:174 2014.

30. Hamani M, Yenikoye A, Banoin M: Some data on sperm from white Fulani and Tuareg rams from Niger. Faculty of Agronomy, University Abdou Niamey (Niger), 1987.

31. Colas G: Seasonal variation in sperm quality in Ile-de-France rams. I. Study of cell morphology and mass motility. *Repr Nutr Dev,* 20 (6): 1789-1799, 1980.

32. Ghozlane F, Ziki B, Yakhlef H: Seasonal variations in quantitative traits of Ouled Djellal ram semen. *Renc Rech Rum*, 12:164, 2005.

33. Benia AR, Taibi K, Ait-Amrane A, Hammoudi SM, Kaidi R: Study of seasonal variations in sexual activity in Algerian rams: Sexual behavior, control of testosterone concentration and environmental factors. *Afr J Biotech,* 12 (41): 6042-6048, 2013.

34. Chemineau P, Gognie Y, Thimonier J: Controlling the reproduction of domestic mammals. **In,** Reproduction in mammals and humans, INRA, Versailles, France, 792-815, 2001.

35. Taherti M, Kaidi R: Productivity of the OuledDjellal ewe according to the mode of breeding. *J Scientist Lebanese*, 19 (1): 47-58, 2018.

36. Schanbacher BD, Wu W, Nienaber JA, Hahn GL: Twenty-four-hour prolactin and testosterone profiles in ram lambs exposed to skeletal photoperiods consisting of various light pulses. *J Reprod Fertil*, 40, 59-68, 1985. DOI: 10.1530/jrf.0.0730037

37. Thiery JC, Chemineau P, Hernandez X, Migaud M, Malpaux B: Neuroendocrine interactions and seasonality. *Domest Anim Endocrinol*, 23, 87-100, 2002. DOI: 10.1016/s0739-7240(02)00148-0

38. Amir D, Volcani R: Seasonal fluctuations in sexual activity of German Awassi Merino, Corriedale, Border-Leicester and Dorset Horn sheep rams. II. Seasonal changes in seminal characteristics. *J Anim Sci*, 64 (1): 121-125, 2009. DOI: 10.1017/S002185960001056X

39. Wilson PR, Lapwwood KR: Studies of hormonal secretion in Romney rams: plasma profiles of luteinizing hormone, testosterone and prolactin, LH/testosterone interrelations and influence of the season. *Theriogenology*,

9 (3): 279-294, 1978. DOI: 10.1016/0093-691X(78)90036-5

40. Gomes WR, Joyce MC: Seasonal changes in serum testosterone in adult rams. *J Anim Sci*, 41, 1375-1375, 1975. DOI: 10.2527/jas1975.4151373x

41. Beltrán-Frutos E, Seco-Rovira V, Martínez-Hernández J, Ferrer C, Serrano-Sánchez MI, Pasteur LM: Cellular changes in spermatogenesis during seasonal testicular regression: An update review in mammals. *Animals*, 12:1605, 2022. DOI: 10.3390/ani12131605

42. Pelletier J, Garnier DH, D e Reviers MM, Terqui M., Ortavant R: Seasonal variations in LH and testosterone release in rams of two breeds. *J Reprod Fertil*, 64, 341-346, 1982. DOI: 10.1530/jrf.0.0640341

43. Zarazaga LÁ, Gatica MC, De La Rosa I, Delgado-Pertíñez M, Guzmán JL: The high testosterone concentrations of the bucks used in the "Male Effect" Is not a prerequisite for obtaining high ovarian activity in goats from Mediterranean latitudes. *Animals*, 12 (8):954, 2022. DOI: 10.3390/ani12080954

44. Maquivar MG, Smith SM, Busboom JR: Reproductive management of rams and ram lambs during the pre-breeding season in US sheep farms. *Animals (Basel),* 11 (9):2503, 2021. DOI: 10.3390/ani11092503

45. Sawyer G, Jitik Naravan E: A review of the influence of climate change on sheep reproduction. *Compar Endocrinol Anim,* 2019. DOI: 10.5772/intechopen.86799

46. Đuričić D, Dobos A, Grbavac J, Stiles C, Bacan I, Vidas Ž, Marković F, Kočila P, Samardžija M: Climatic impacts on reproductive performance of Romanov sheep in temperate climate. *J Anim Biometeorol Behavior,* 10 (1): 2201-2022, 2021. DOI: 10.31893/jabb.22001