



## RESEARCH ARTICLE

### Seasonal Variations in Semen Characteristics in Arabic Rams

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#### ABSTRACT

The aim of this study was to evaluate the seasonal variations in semen characteristics in Arabic rams. 8 adult Arabic rams at the age of 2-3 years were used for this research. Semen was collected with electro ejaculator every 15 days for a period of 6 months (3 months of in breeding season and 3 months of non breeding season). Semen samples of these rams were subjected to the parameters including semen volume, sperm motility, sperm concentration, abnormal and live sperm percentage. Semen quality difference between breeding and non breeding seasons was significant. Semen volume, sperm motility, percent of live sperm, the percentage of abnormal sperm and sperm concentration determined in breeding and non breeding seasons had significant differences. Therefore, better progressive motility of sperm, sperm concentration, percent of live sperm and low percentage of abnormal sperm clearly explained high fertility potential of male in breeding season when compared with non breeding season. In conclusion, this study indicated that sexual performance of Arabic rams completely dependent on season.

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#### INTRODUCTION

Although ram can copulate with ewe throughout the year, but quantitative and qualitative decrease in semen production and sperm fertility during the non breeding season has been reported (Corteel, 1977; Colas, 1980). Photoperiod or annual season is the main environmental factor controlling the seasonality of reproduction (Hafez, 2000; Kaya *et al.*, 2000). Photoperiod or annual season may influence reproductive physiology and fertilization potential of male by change in hormonal profile and histological variations of testis cells in various seasons (Alkass *et al.*, 1982; Hamidi *et al.*, 2010a). However, other environmental stimulators including availability of food and social interactions should not be disregarded as potential regulators of the sensuality of reproduction performance (Walkden-Brown *et al.*, 1993; Mani *et al.*, 1996). The present study was performed to assess seasonal variations in semen characteristics in Arabic rams during breeding and non breeding seasons.

#### MATERIALS AND METHODS

The study was carried out with 8 Arabic rams aging 2 to 3 years. Experiments were performed in research farm

of Ramin Agriculture University in Khuzestan province in Iran located at latitude 31° 52', where October to December is included as breeding season and January, February and early days of March as non breeding season. The frequency of semen collection was twice per month for a period of 6 months (3 months of breeding season and 3 months of non breeding season) with electro ejaculator. Semen samples were evaluated on the basis of following parameters: semen volume, motility percentage, concentration, abnormal sperm and live sperm percentage.

**Semen evaluation:** Semen samples were evaluated using standard methods (Hafez, 2000). The ejaculated semen volume was measured in graduated tube. The live sperm percent was determined using eosin-nigrosin staining technique by counting 100 spermatozoa under oil immersion objective (x 1000). The abnormal sperm percent was determined by evaluation a total of 300 spermatozoa from eosin-nigrosin stained smears. Sperm concentration per milliliter was determined with a hemocytometer after diluting semen with a 0.05% formaldehyde saline solution (1:400) and examining under microscope (x 400).

Sperm motility was assessed subjectively under a phase contrast microscope at x 400 magnification. Noted to the high correlation between sperm motility and

fertility rate, this parameter of spermatozoa traits highly and carefully subjected by computer aided sperm analyzer (CASA) and following criteria were evaluation in these procedures: curvilinear velocity of sperm (CLV), average path velocity of sperm (APV), velocity of straight line (VSL) and linearity of sperm motility (LIN). Based on sperm CASA, the sperm motility was classified in four classes included: fast progressive (Class A), moderate progressive (Class B), non progressive (Class C), less motile (Class D).

**Statistic analysis:** Semen volume, sperm motility, sperm concentration, percent of abnormal sperm, percent of live sperm percent, curvilinear velocity, average path velocity, velocity of straight line and linearity of sperm motility in two seasons (breeding and non breeding) were analyzed by paired-sample t test in SPSS program.

## RESULTS

Investigation of semen quantity and quality in breeding and non breeding seasons showed significant differences. Semen volume, sperm motility, percent of live sperm, percentage of abnormal sperm and sperm concentration per milliliter semen determined in breeding and non breeding seasons were significantly different ( $P<0.01$ ) (Table 1).

Among various motility classes of spermatozoa in breeding and non breeding seasons significant differences were observed in CASA program (Table 2). The comparison of sperm motility characteristics between breeding and non breeding season by CASA program is presented in Table 3. Analysis of motility characteristics of sperm by CASA program showed significant differences in CLV, APV, VSL and LIN of sperms in two seasons ( $P<0.01$ ).

## DISCUSSION

Semen characteristics were significantly different between breeding and non breeding seasons. These differences were observed in semen volume, sperm concentration per milliliter, percentage of live sperm, abnormal sperm and sperm motility. High semen volume was noted in breeding season and its value decreased in beginning the non breeding season (Table 1). Sperm concentration also demonstrated significant change between breeding and non breeding seasons, in which high sperm concentration was showed in breeding season (Table 1). The presented findings were in agreement with results that previously reported for seasonal variations in semen volume and sperm concentration of ram and buck (Zarazaga *et al.*, 2009; Suhair and Abdolla, 2010). Similar to our results, Gündoğan (2007) observed that ejaculate volume and sperm concentration in Chios and five Daglic fat-tailed rams were higher during the autumn (breeding season) than the other months of the year. Decrease in semen volume and sperm concentration can be attributed to the decreased function of pituitary gland and testis due to decrease in their sizes (Hamidi *et al.*, 2010a) in the non breeding season. Hamidi *et al.* (2010a) reported that the concentrations of hormones released from pituitary gland

**Table 1:** Seasonally variation of subjecting parameters

Parameters	Breeding season	Non breeding season
Semen volume (ml)	1.65±0.10**	1.05±0.04
Sperm motility (%)	84.8±0.7**	74.8±0.7
Sperm concentration( $\times 10^6$ ml <sup>-1</sup> )	2920±0.1**	2610±0.1
Live sperm percent (%)	4.7±0.4**	79.0±0.9
Abnormal sperm percent (%)	4.7±0.4**	7.2±0.3

Data are presented as Mean±SEM; \*\* Differences of means in each row significant at  $P<0.01$ .

**Table 2:** Seasonal variation of spermatozoa motility classes

Spermatozoa motility classes	Breeding season	Non breeding season
(A)	59.4±1.2**	39.3±1.7
(B)	26.4±0.9**	21.6±0.9
(C)	20.7±0.9**	11.9±1.0
(D)	13.4±1.3**	7.4±1.0

Data are presented as Mean±SEM; \*\* Differences of means in each row significant at  $P<0.01$ .

**Table 3:** The comparison of sperm motility characteristics between breeding and non breeding season by CASA program (No abbreviation in title)

Sperm motility Classes	Motility parameters	Breeding season	Non breeding season
A	APV ( $\mu$ m/s)	91.6±1.4**	71.0±2.4
	VSL ( $\mu$ m/s)	79.2±2.7**	44.2±2.1
	VCL ( $\mu$ m/s)	129.8±2.9**	91.0±2.5
	LIN (%)	59.8±1.2**	48.5±1.0
B	APV ( $\mu$ m/s)	45.0±1.2**	33.4±0.7
	VSL ( $\mu$ m/s)	31.6±0.9**	18.1±0.7
	VCL ( $\mu$ m/s)	76.7±1.3**	61.8±0.9
	LIN (%)	41.3±1.4**	29.2±0.9
C	APV ( $\mu$ m/s)	18.9±0.8**	10.8±0.5
	VSL ( $\mu$ m/s)	10.3±0.3**	4.5±0.2
	VCL ( $\mu$ m/s)	72.4±1.2**	50.2±1.9
	LIN (%)	14.3±0.6**	8.9±0.2
D	APV ( $\mu$ m/s)	5.6±0.5**	2.5±0.2
	VSL ( $\mu$ m/s)	1.9±0.1**	0.6±0.1
	VCL ( $\mu$ m/s)	27.1±1.4**	13.8±0.4
	LIN (%)	7.2±0.4**	4.6±0.4

\*\*The means in same rows differed significantly at  $P<0.01$ .

and testis of Arabic rams significantly decreased in the non breeding season. Also, Dorostgoal (2006) reported that histological and histometrical structure of seminiferous tubules in adults Arabic rams significantly changed in various seasons and this could severely influence sex performance of male.

Evaluation of the spermatozoa viability demonstrated the significant differences in percent of live spermatozoa between breeding and non breeding seasons (Table 1). The percentage of live sperms was 87.36% for the breeding season and 79% in the non breeding season. Moreover, between breeding and non breeding seasons, significant difference was observed in percentage of abnormal sperm, in which higher percentage of abnormal sperms was noted in non breeding season than breeding season (Table 1). Suhair and Abdalla (2010) reported that percentage of live and abnormal sperms in semen of Desert rams showed significantly changes in various seasons. Gündoğan (2007) reported a moderate decrease of abnormal sperm in breeding season (autumn) as compared to the non breeding season in Daglic fat-tailed rams. Galil and Galil (1982) and Suhair and Abdalla (2010) reported seasonal change in percentage of live and abnormal sperms in Desert rams. Decreased live sperm percentage and an increase in a percentage of abnormal sperm in non breeding season could be related to the

decrease in number of seminiferous tubules and increase in degenerative processes (Suhair and Abdalla, 2010). Structural variations have been reported in the seminiferous epithelium, with affected spermatogenic and sertoli cells by Dorostgoal (2006). The height of germinal epithelium, spermatogenic cell density, seminiferous tubule diameter, number and size of sertoli cells in Arabic ram were significantly different in various seasons of the year (Dorostgoal, 2006). This may severely affected the sexual performance and spermatogenic process of male and presumably influence percentage of live and abnormal sperm. Sertoli cells are responsible for the blood-testis barrier and facilitated nourishment of sexual germ cells in male (Hafez, 2000).

Several studies have been shown a high correlation between sperm motility and fertility (Farrell *et al.*, 1998). Sperm motion parameters are very important for the passage of sperm from various barriers in reproductive female tract and penetration in to the oocyte (Cox *et al.*, 2006). Analysis of sperm motility parameters using CASA indicated interesting results (Tables 2-4). Sperm motility percentage of various classes in breeding and non breeding seasons was significantly different. High motion of sperms in various classes was showed in the breeding season as compared to the non breeding season (Table 2). The changes of CLV, APV, VSL and LIN in various classes and in two seasons were reported in this paper. The tables can be helpful to explain the causes of high fertility potential of male in breeding season. This study showed significant differences among the above said sperm motility classes (Table 4). In the non breeding season, percentage of sperm motility, linearity and velocity of sperm motion decreased significantly when compared with breeding season ( $P < 0.01$ ). The changes of sperm motion traits in the different seasons of the year previously have been reported for many breeds of rams and buck. Zarazaga *et al.* (2009) reported significant differences in the percentage of sperm motility and sperm motion traits of goats in various seasons. Suhair and Abdalla (2010) reported significant differences in sperm motility percentage of Desert rams determined in various seasons. Seasonal variations of progressive sperm motility in local crossbred and Merino rams have also been reported by Saleh (1997) and Fowler (1965), respectively. The current study revealed significantly ( $P < 0.01$ ) lower sperm motility values during non breeding season as compared to the breeding season. This response can be caused by different factors, including availability of essential nutrients in various seasons (Amir and Volcani, 1965; Onstad, 1967), seasonal variations of hormonal profile (Dorostgoal, 2006; Hamidi *et al.*, 2010b), structural variations of leydig and sertoli cells in breeding and non breeding seasons (Dorostgoal, 2006) and other factors. Endocrine system and their hormonal profile especially male sex hormone have definite and important roles in spermatogenesis and sperm maturations (Johnson *et al.*, 1973; Sanford *et al.*, 1974; Gomes and Joyce, 1975). Love level of this hormone was determined in the non breeding season as compared to the breeding season in Arabic rams (Dorostgoal, 2006; Hamidi *et al.*, 2010a). Moreover, Dorostgoal (2006) reported that the size and number of Leydig cells in the testis, responsible for

synthesis and release of testosterone hormone exhibit significant changes in various seasons. High number and size of these cells were shown in the breeding season as compared to the non breeding season. Testosterone hormone plays an important role in maturation and capacitation of spermatozoa and increase progressive sperm motility outside the testis (Hafez, 2000). Hence, high progressive motility of sperm, sperm concentration, live sperm percentage and low percentage of abnormal sperm clearly explain the high fertility potential of male in breeding season as compared to the non breeding season. In conclusion, this study indicated that sexual performance of Arabic rams is completely dependent on the change of season, and their fertility potential showed seasonal variations.

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