The Effects of Feeding Lucerne Hay on Semen Characteristics in Desert Rams under Seasonal Thermal Environment

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Abstract

The present study involved nine entire Desert rams (Hamari) to investigate the effects of the level of feeding lucerne hay (High, Medium and Low) and the possible effects of the seasons (winter and summer). The studied parameters included the scrotal circumference (Sc), rectal temperature (Tr) and semen characteristics. Feeding low level of lucerne hay significantly lowered Sc and Tr (morning and evening) in both seasons compared to medium and high levels. During summer and winter the ejaculate volume (EV) was significantly lower with the low level of feeding compared to the other feeding levels. The sperm mass motility (SMM) was significantly lower with the medium and low levels of feeding compared to the higher level during summer. The percentage of sperm individual motility (SIM) was significantly lower with the medium and low levels of feeding during summer and winter. However, the sperm cell concentration (SCC) significantly increased during winter and summer with medium and low levels of feeding respectively compared to the higher level. The abnormal sperm percent (ASP) was significantly higher with the low level of feeding during summer compared to the respective groups. Summer season significantly increased the afternoon Tr values of all feeding groups compared to winter values. All feeding levels had a significantly lower EV, SMM, SIM and a significantly higher ASP values during summer compared to winter values. It could be concluded that seminal trait of Sudanese Desert rams were influenced by feed restriction, seasonal change and heat stress of thermal environment.

Introduction

The domestic sheep (*Ovis aries*) are widely distributed in Sudan, they are characterized by their high adaptive capabilities to different climatic conditions. Their population is estimated at 47 million; Desert sheep constitute more than 60% of sheep population and they are strictly confined to desert and semi-desert zones in northern Sudan (5). As sheep in Sudan have contribution to national economy, it is worth studying factors that might retard their productive and reproductive potentials .This study is designed to investigate the effects of feeding three levels of lucerne hay during summer and winter on seminal traits of Desert rams.

Materials and Methods

Nine entire adult apparently healthy Hamari Desert rams aged 2-3 years and weighing 39.0-46.5 kg were used in the experiment .They were randomly assigned to groups of 3 rams. each group fed on high, medium and low level of sun-dried lucerne hay (*Medicago sativa*).They were accommodated in separate pens; clean and fresh tap-water was continually offered. The composition of the feeds is presented in Table (1).

Table1.The chemical composition of the feeds (%) on dry matter basis .

	Lucerne (*Medicago sativa*)
Dry matter	23
Metabolizable energy(MJ/kg)	8.48
Crude fibre (%)	30.00
Crude protein (%)	17.5
Ether extract (%)	0.99
Nitrogen free extract (%)	40.27
Ash (%)	11.57

A certified mercury-in-glass clinical thermometer was used to measure rectal temperature (Tr), the body weight (BW) was measured by a spring balance (Salter No.235-Trade No.2892- England). The scrotal circumference was measured by a tape. Semen samples were collected using the electric ejaculator, Raukura ram design probe (Mark IV OLVET, New Zealand}.

The rams were allowed an adaptive period of 2 weeks; when the considered as the high level of feeding (1200gm), accordingly the medium level 66% (800gm) and the low level, 33%(400gm) were calculated.

The maintenance requirements (Mm) for sheep, kept in doors, was calculated as Mm/kg= 1.2+0.1 W (body weight) (**26**).The initial average BW of rams obtained was 43.22 kg, accordingly the maintenance allowance was 6.81686 MJ/day. The metabolizable energy (ME) of lucerne hay in ruminants was calculated as 8.80 MJ/kg (**36**). The amount of lucerne needed for maintenance requirements was computed as 774.84 gm. Therefore the medium level of feeding used in this experiment could provide approximately the maintenance requirements and the low level imposed was below maintenance. During summer and winter, the same protocol of feeding regimen was applied.

The measurements of (Tr) were performed twice daily, in the morning (7:00 a.m.) and in the afternoon (2:00 p.m.), to the nearest ± 0.1 °C, during summer and winter for 12 weeks. The scrotal circumference was measured weekly to the nearest ± 0.1 cm. In each season, for 10 weeks, semen samples were collected weekly at 8:00 am, by electro-ejaculation according to the method described by (**7**) and analyzed according to the method adopted by (**9 and 10**) and (**6**). The experimental protocol is shown in table (2).

Table 2. Experimental protocol

Experimental period	Treatment	Daily measurements	Weekly sampling and measurements	Analysis
Winter (December/99-March 2000)	High, medium and low levels of feeding	Tr (7:00 a.m.) Tr (2:00 p.m.)	Semen Scrotal circumference	Semen characteristics
Summer (June – September 2000)	High, medium and low levels of feeding	Tr (7:00 a.m) Tr (2:00 p.m)	Semen Scrotal circumference	Semen characteristics

Statistical Analysis

Recollected data were statistically analyzed according to Statistical Analysis System (**34**); the Analysis of variance (ANOV A) test was also used.

Results and Discussion

The results data obtained are presented as means ± S.E. The prevailing climatic conditions during experimental period are presented in Table (3).

During both seasons, the rams exhibited significant [($p<0.05$) and ($p<0.01$)] lower Tr with low level of feeding in the morning (Table, 4) and afternoon (Table, 5) respectively, compared to values obtained for the other groups. These results were attributed to the decreased metabolic heat production in response to feed restriction (**8; 19; 31 and 16**) .The present findings are in agreement with that reported by (**1**). The significantly ($p<0.01$) higher Tr values obtained in the afternoon for all feeding levels during summer compared to winter is attributed to the increase in thermal load with rise of ambient temperature.

Table (6) Showed that, in both seasons, feed restriction significantly ($p<0.01$) lowered SC. However, the rams maintained on high level of feeding had significantly ($p<0.01$) higher values of SC during winter compared to summer values. The reduction in SC was attributed to the general loss in the mean BW (Table, 14) and loss of subcutaneous fat in the scrotal sac. Similar findings were reported in feed restricted rams by (**2; 25; 15 and 30**).

The higher SC was recorded for high level of feeding during winter compared to summer values (Table, 6). This might be attributed to the increase in the number of seminiferous tubules and spermatogenic activity. Similarly, (3) reported relatively higher values of SC during winter conditions in Desert rams. In temperate zone during cold season, (**22**) reported higher values of SC of Finnish Land Race and Tasmanian rams.

The EV was significantly ($p<0.01$) lower with low level of feeding in both seasons (Table, 7). The low response was apparently associated with feed restriction and low concentration of luteinizing hormone (LH) and reduction of testosterone secretion (**11**). The low EV could be attributed to the decreased function of pituitary gland and testis due to decrease in their sizes in feed restricted rams (**2**). The significantly higher EV was observed during winter in all rams in comparison to that summer (Table, 7). The findings could be related to the effects of endocrine activation on exposure to cold and stimulatory effects of testosterone on accessory genital glands.

The current findings are in agreement with the findings reported by (**17**) and (**3**) in Desert rams.

The SMM was significantly ($p<0.05$) lower during summer with feed restriction (Table, 8). This is clearly related to decline in nutritional status of rams. The low food intake during summer could have induced low fructose level in seminal plasma and consequently decreased SMM as they are positively related (**4**). Moreover, (**18**) recorded a low level of fructose in seminal plasma of Desert rams during summer and they attributed these results to high ambient temperature and reduced secretion of testosterone. Additionally, (**12**) attributed the reduction of SMM during summer to low fructose level and sperm metabolic rate. The reduction of SMM could be partly associated with low serum albumin concentration in feed restricted rams (Table, 13). (**29**) suggested that serum albumin is the best medium for increasing sperm motility in vitro.

In agreement with (**18**), the significantly ($p<0.01$) lower SMM observed with all levels of feeding during summer compared to winter values (Table, 8) might be attributed to high ambient temperature prevailing consequently high Tr of rams which recorded during summer.

Table (9) showed that, the percentage of SIM was significantly lower during winter ($p<0.01$) and summer ($p<0.05$) with the medium and low levels of feeding. This might be mainly due to low plane of nutrition and protein intake resulting in a relatively low concentration of seminal plasma metabolites associated with thermal stress during summer. (**14**) attributed the low SIM to decline in sex hormones binding globulins. Similarly (**35**) reported that the reduced SIM in feed restricted rams was attributed to low seminal plasma fructose concentration and depressed activity of the pituitary gland.

Irrespective of level of feeding, SIM was significantly ($p>0.001$) higher during winter compared to summer values (Table, 9).This could be attributed to efficient testicular thermoregulation mechanism.

Rams maintained on medium level of feeding showed a significantly ($p<0.05$) higher SCC during winter (Table, 10). This response could be associated with the low EV obtained with feed restriction and the fact that EV depends primarily upon the secretion of seminal plasma rather than SCC (**20**). However, the significantly ($p<0.05$) lower SCC reported during summer with the medium level of feeding could be associated with the increase in ambient temperature. These findings come in consistence with that reported by (**32 and 2**).

The SCC was lower with all levels of feeding during summer compared to winter (Table, 10). However, the decrease was significant ($p<0.001$) in rams maintained on medium level of feeding. The obtained data were attributed to decrease in spermatogenic activity and epididymal reserve during the hot season due to spermatogenic degeneration. Similarly a lower SCC was reported in Desert rams during summer compared to winter (**18**).

The current study revealed a significant ($p<0.05$) lower LSP values obtained during summer compared to winter in rams maintained on medium level of feeding (Table, 11) The findings could be related to the increase in body temperature resulting in increase in metabolic activity of testicular cells leading to testicular hypoxia which probably plays a role in heat-induced spermatogenic disturbance (**12**) and/or attributed to the reduction in number of seminiferous tubules and increase in dead sperm percent due to increase in degenerative process (**17**).

Feed restriction significantly ($p<0.05$) increased the incidence of ASP during summer (Table, 12).). Similar findings were reported by (**37; 11 and 38**). This response might be due to the decrease in availability and supply of essential nutrients required for sperm production in the testis and epididymal maturation associated with thermal stress during summer (**35 and 13**).

The ASP was higher during summer with all levels of feeding compared to winter, (Table, 12); the value was significant ($p<0.05$) with the ($p<0.001$) high, medium and low levels of feeding. The results explained that the high body temperature reported during summer and impairment of testicular cooling efficiency (**33**) and consequently the increased testicular temperature provokes testicular degeneration (**27; 28; 23; 21;24 and 18**).

It could be concluded that seminal trait of Sudanese Desert rams were influenced by feed restriction, seasonal change and heat stress of thermal environment

Table 3. The mean values of ambient temperature Ta (°C) and relative humidity, RH (%) during the experimental period.

Season	Ta (°C)			RH (%)
	Max.	Min.	Mean	(Mean)
Winter (December/99 -March/ 2000)	31.58 ± 1.51	15.83 ± 1.66	23.71 ± 7.88	28.17 ± 4.57
Summ^r (June - September /2000)	38.23 ± 2.01	25.59 ± 1.18	31.91 ± 6.32	42.09 ± 11.49

Table 4. Effects of level of feeding lucerne hay and season on rectal temperature, Tr (°C) of Desert rams at 7:00 a.m. (n = 252, mean ± S.E).

Level of feeding	Season: Winter	Season: Summer	SL
High	$^{A}37.98^{a}$ ±0.04	$^{A}38.12^{a}$ ±0.02	NS
Medium	$^{A}37.74^{a}$ ±0.05	$^{B}37.89^{a}$ ±0.04	NS
Low	$^{B}37.40^{a}$ ±0.06	$^{B}37.54^{a}$ ±0.10	NS
SL	*	*	

. Mean values within the same row bearing similar superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level.
* Significant at $p<0.05$.
NS: Not significant

Table 5. Effects of level of feeding lucerne hay and season on rectal temperature, Tr (°C) of Desert rams at 2:00 p.m. (n = 252, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}38.72^{b}$ ± 0.03	$^{A}39.10^{a}$ ± 0.05	**
Medium	$^{A}38.64^{b}$ ± 0.04	$^{A}38.99^{a}$ ± 0.06	**
Low	$^{B}38.39^{b}$ ± 0.04	$^{B}38.76^{a}$ ± 0.05	**
SL	**	**	

. Mean values within the same row bearing different superscripts (small) were non significantly different.

. Mean values within the same column bearing different superscripts (capital) were significantly different.

SL: Significance level

* *: Significant at $p< 0.01$.

NS: Not significant

Table 6. Effects of level of feeding lucerne hay and season on scrotal circumference, SC (cm) in Desert rams.

(n = 30, mean ± S.E.).

Level of feeding	Season: Winter	Season: Summer	SL
High	$^{A}32.52^{b}$ ±0.40	$^{A}30.75^{a}$ ±0.19	**
Medium	$^{B}30.55^{a}$ ±0.35	$^{AB}29.72^{a}$ ±0.42	NS
Low	$^{B}27.77^{a}$ ±0.51	$^{B}28.77^{a}$ ±0.45	NS
SL	**	**	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
**: Significant at p< 0.0 1.
NS: Not significant

Table 7. Effects of level of feeding lucerne hay and season on ejaculate volume, EV (ml) in Desert rams.
(n = 30, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}1.96^{a}$ ± 0.09	$^{A}1.66^{b}$ ± 0.09	**
Medium	$^{A}1.98^{a}$ ± 0.10	$^{A}1.58^{b}$ ± 0.09	**
Low	$^{B}1.58^{a}$ ± 0.08	$^{B}1.33^{b}$ ± 0.07	*
SL	**	**	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at p< 0.05.
**: Significant at p< 0.0 1.
NS: Not significant

Table 8. Effects of level of feeding lucerne hay and season on sperm mass motility, S.M.M (0 - 5) in Desert rams.

(n = 30, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}4.65^{a}$ ± 0.08	$^{A}3.66^{b}$ ± 1.41	***
Medium	$^{A}4.42^{a}$ ± 0.08	$^{B}2.86^{b}$ ± 1.51	***
Low	$^{A}4.32^{a}$ ± 0.10	$^{B}3.02^{b}$ ± 1.34	***
SL	NS	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at p< 0.05.
**: Significant at p< 0.0 1.
NS: Not significant

Table 9. Effects of level of feeding lucerne hay and season on sperm individual motility, S IM (%) in Desert rams.

(n = 30, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	[A] 76.67 [a] ± 2.24	[A] 59.14 [b] ± 4.26	**
Medium	[B] 68.50 [a] ± 2.90	[B] 30.71 [b] ± 4.71	**
Low	[B] 63.33 [a] ± 2.84	[B] 35.15 [b] ± 4.69	**
SL	**	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at $p< 0.05$.
**: Significant at $p< 0.01$.
NS: Not significant

Table 10 Effects of level of feeding lucerne hay and season on sperm cell,SCC concentration ($\times 10^9$/ml)in Desert rams.

(n = 30, mean ± S.E..).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{B}2.07^{a}$ ± 0.133	$^{AB}1.64^{a}$ ± 0.21	NS
Medium	$^{A}2.37^{a}$ ± 0.15	$^{B}1.24^{b}$ ± 0.13	***
Low	$^{B}2.15^{a}$ ± 0.21	$^{A}1.80^{b}$ ± 0.21	NS
SL	*	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at $p< 0.05$.
* *: Significant at $p< 0.01$.
NS: Not significant

Table 11. Effects of level of feeding lucerne hay and season on the incidence of live sperm (%), LSP in Desert rams.

(n = 30, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}94.80^{a}$ ± 1.37	$^{A}93.24^{a}$ ± 3.40	NS
Medium	$^{A}94.97^{a}$ ± 0.82	$^{B}87.60^{b}$ ± 3.37	*
Low	$^{A}94.55^{a}$ ± 1.00	$^{A}91.28^{a}$ ± 2.14	NS
SL	NS	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at $p < 0.05$.
**: Significant at $p < 0.01$.
NS: Not significant

Table12. Effects of level of feeding lucerne hay and season on the incidence of abnormal sperm (%), ASP in Desert rams.

(n = 30, mean ± S.E.).

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}3.18^{b}$ ± 0.60	$^{B}13.07^{a}$ ± 4.07	*
Medium	$^{A}5.45^{b}$ ± 1.22	$^{B}13.36^{a}$ ± 3.29	*
Low	$^{A}3.86^{b}$ ± 1.11	$^{A}21.18^{a}$ ± 4.03	**
SL	NS	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at $p < 0.05$.
**: Significant at $p < 0.01$.
NS: Not significant

Table 13. Effects of level of feeding Lucerne hay and season on serum albumin concentration (g/dl) in Desert rams.

(n = 36, mean ± S.E.)

Level of feeding	Season		SL
	Winter	Summer	
High	$^{A}3.57^{a}$ ± 0.04	$^{A}3.45^{b}$ ± 0.04	*
Medium	$^{B}3.50$ ± 0.04^{a}	$^{B}3.37^{b}$ ± 0.05	*
Low	$^{AB}3.54^{a}$ ± 0.04	$^{B}3.38^{b}$ ± 0.05	*
SL	*	*	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at $p < 0.05$.
* *: Significant at $p < 0.0 1$.
NS: Not significant

Table 14 Effects of level of feeding lucerne hay and season on the mean body weight, BW (kg) of Desert rams.

(n = 36, mean ± S.E.M.)

Level of feeding	Season		SL
	Winter	Summer	
High	^{A}44.16^{a} ±0.33	^{A}42.16^{b} ±0.40	***
Medium	^{B}39.57 ±0.40^{a}	^{B}38.40^{b} ±0.37	***
Low	^{B}35.70^{a} ±0.39	^{B}34.45^{b} ±0.60	***
SL	**	***	

. Mean values within the same row bearing different superscripts (small) were non significantly different.
. Mean values within the same column bearing different superscripts (capital) were significantly different.
SL: Significance level
* : Significant at p< 0.05.
* *: Significant at p< 0.0 1.
NS: Not significant

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