

Table S1. Metabolisable energy (ME), metabolisable protein (MP) and mineral balances in high 10%, control group and low 10% at 2-month-old lambs (start experiment).

	ME (Mcal/day)	MP (g/day)	Ca (g/day)	P (g/day)
High 10%				
Intake	3.044	117	11	4
Total requirement	3.044	95	8	4
Maintenance	1.066	49	3	2
Maintenance (cold stress)	0.000	-	-	-
Growth	1.977	46	5	2
Balance	0.000	21	3	0
Control group				
Intake	2.787	116	8	4
Total requirement	2.787	90	8	4
Maintenance	1.071	49	3	2
Maintenance (cold stress)	0.000	-	-	-
Growth	1.716	41	4	2
Balance	0.000	26	1	0
Low 10%				
Intake	2.506	115	9	6
Total requirement	2.506	83	7	4
Maintenance	1.070	49	3	2
Maintenance (cold stress)	0.000	-	-	-
Growth	1.436	35	4	2
Balance	0.000	25	2	2

Table S2. Ration summary balances in high 10%, control group and low 10% at 2-month-old lambs (start experiment).

Feed name	Dry matter Fed (kg/day)	As Fed (kg/day)	Total (%)
High 10%			
Alfalfa Hay – Medium Bloom (204) Medium Chop	0.330	0.363	27.50
Wheat - Straw (320) Medium Chop	0.140	0.157	11.67
Barley Grain - Heavy (401) Finely Ground	0.600	0.682	50.00
Wheat Bran - (620) Finely Ground	0.090	0.101	7.50
Calcium - Carbonate (805)	0.005	0.005	0.42
Salt (831)	0.005	0.005	0.42
Megalac	0.030	0.031	2.50
Total	1.200	1.343	100.00
Control group			
Alfalfa Hay - Medium Bloom (204) Medium Chop	0.400	0.440	33.33
Wheat - Straw (320) Medium Chop	0.200	0.225	16.67
Barley Grain - Heavy (401) Finely Ground	0.500	0.568	41.67
Wheat Bran - (620) Finely Ground	0.090	0.101	7.50
Calcium - Carbonate (805)	0.005	0.005	0.42
Salt (831)	0.005	0.005	0.42
Total	1.200	1.344	100.00
Low 10%			
Alfalfa Hay - Medium Bloom (204) Medium Chop	0.410	0.451	34.17
Wheat - Straw (320) Medium Chop	0.290	0.326	24.17
Barley Grain - Heavy (401) Finely Ground	0.210	0.239	17.50
Wheat Bran - (620) Finely Ground	0.280	0.315	23.33
Calcium - Carbonate (805)	0.005	0.005	0.42
Salt (831)	0.005	0.005	0.42
Total	1.200	1.340	100.00

Table S3. Metabolisable energy (ME), metabolisable protein (MP) and mineral balances in high 10%, control group and low 10% at 9-month-old lambs (End experiment).

	ME	MP	Ca	P
	(Mcal/day)		(g/day)	
High 10%				
Intake	4.912	186	18	7
Total requirement	4.912	115	12	6
Maintenance	1.888	71	6	3
Maintenance (cold stress)	0.000	-	-	-
Growth	3.024	44	6	3
Balance	0.000	71	5	0
Control group				
Intake	4.498	184	13	6
Total requirement	4.498	109	11	6
Maintenance	1.888	71	6	3
Maintenance (cold stress)	0.000	-	-	-
Growth	2.610	38	6	2
Balance	0.000	75	2	1
Low 10%				
Intake	4.083	183	14	9
Total requirement	4.083	102	10	5
Maintenance	1.924	71	6	3
Maintenance (cold stress)	0.000	-	-	-
Growth	2.159	32	5	2
Balance	0.000	70	3	4

Table S4. Ration summary balances in high 10%, control group and low 10% at 9-month-old lambs (end of experiment).

Feed name	Dry matter Fed (kg/day)	As Fed (kg/day)	Total (%)
High 10%			
Alfalfa Hay - Medium Bloom (204) Medium Chop	0.540	0.593	28.13
Wheat - Straw (320) Medium Chop	0.220	0.247	11.46
Barley Grain - Heavy (401) Finely Ground	0.950	1.080	49.48
Wheat Bran - (620) Finely Ground	0.144	0.162	7.50
Calcium - Carbonate (805)	0.008	0.008	0.42
Salt (831)	0.008	0.008	0.42
Megalac	0.050	0.051	2.60
Total	1.920	2.149	100.00
Control group			
Alfalfa Hay - Medium Bloom (204) Medium Chop	0.640	0.703	33.33
Wheat - Straw (320) Medium Chop	0.320	0.360	16.67
Barley Grain - Heavy (401) Finely Ground	0.800	0.909	41.67
Wheat Bran - (620) Finely Ground	0.144	0.162	7.50
Calcium - Carbonate (805)	0.008	0.008	0.42
Salt (831)	0.008	0.008	0.42
Megalac	0.000	0.000	0.00
Total	1.920	2.150	100.00
Low 10%			
Alfalfa Hay - Medium Bloom (204) Medium Chop	0.650	0.714	33.85
Wheat - Straw (320) Medium Chop	0.460	0.517	23.96
Barley Grain - Heavy (401) Finely Ground	0.335	0.381	17.45
Wheat Bran - (620) Finely Ground	0.460	0.517	23.96
Calcium - Carbonate (805)	0.008	0.008	0.42
Salt (831)	0.007	0.007	0.36
Megalac	0.000	0.000	0.00
Total	1.920	2.144	100.00

Table S5. Sexual behavior activities of Ghezel lambs with different treatments.

Age [1]	Trait Treat	Foot stamping/kicking			Ano-genital sniffing			Noising			Flehmen			Mount attempts			Mounts			Mounts with ejaculation		
		M	SEM	p	M	SEM	p	M	SEM	p	M	SEM	p	M	SEM	p	M	SEM	p	M ^[2]	SEM ^[3]	p ^[4]
28	Control	0.0	0.0	0.05	0.0	0.0	0.05	0.5 ^b	0.0	0.05	0.0	0.0	0.05	0.0	0.0	0.05	0.0	0.0	0.05	0.0	0.0	0.05
	High	0.0	0.0		0.0	0.0		1.0 ^c	0.2		0.0	0.0		0.0	0.0		0.0	0.0		0.0	0.0	
	Low	0.0	0.0		0.0	0.0		0.0 ^a	0.0		0.0	0.0		0.0	0.0		0.0	0.0		0.0	0.0	
30	Control	2.8 ^b	0.4	0.05	3.1 ^b	0.2	0.05	4.6	0.5	0.05	1.2 ^b	0.1	0.05	0.0 ^a	0.0	0.05	0.0	0.0	0.05	0.0	0.0	0.05
	High	3.3 ^c	0.2		3.5 ^c	0.6		4.0	0.8		2.0 ^c	0.2		2.0 ^b	0.3		0.0	0.0		0.0	0.0	
	Low	0.0 ^a	0.0		1.0 ^a	0.1		4.0	0.4		0.0 ^a	0.0		0.0 ^a	0.0		0.0	0.0		0.0	0.0	NS
32	Control	4.0 ^b	0.9	0.05	6.4 ^b	0.8	0.05	12.0 ^a	1.0	0.05	1.8 ^b	0.3	0.05	1.5 ^b	0.0	0.05	1.0 ^b	0.0	0.05	1.0 ^b	0.0	0.05
	High	6.1 ^c	1.0		8.2 ^c	0.9		12.9 ^{bc}	1.5		3.1 ^c	0.6		2.1 ^c	0.2		1.8 ^c	0.5		1.5 ^c	0.2	
	Low	2.5 ^a	0.3		4.8 ^a	0.4		13.0 ^b	1.6		0.9 ^a	0.0		0.8 ^a	0.2		0.0 ^a	0.0		0.0 ^a	0.0	NS
34	Control	15.3 ^b	2.2	0.05	9.3 ^b	0.7	0.05	16.0	1.3	NS	4.0 ^b	0.2	0.05	3.0 ^b	0.0	0.001	2.0 ^b	0.0	0.01	2.0 ^b	0.1	0.05
	High	18.6 ^c	3.1		10.0 ^b	1.0		15.6	2.1		6.2 ^c	0.5		4.5 ^c	0.1		4.0 ^c	0.5		3.0 ^c	0.4	
	Low	12.0 ^a	3.0		7.0 ^a	0.6		16.0	1.5		2.4 ^a	0.4		1.0 ^a	0.2		1.0 ^a	0.0		0.0 ^a	0.0	NS
36	Control	22.0 ^b	4.8	0.05	12.5 ^b	0.7	0.05	24.0 ^b	2.6	0.05	6.5 ^b	0.9	0.05	8.5 ^b	0.6	0.05	4.5 ^b	0.3	0.05	3.5 ^b	0.3	0.05
	High	24.1 ^c	4.2		14.2 ^c	0.8		23.3 ^b	2.2		7.0 ^b	0.8		9.0 ^b	0.6		4.2 ^b	0.6		3.0 ^b	0.5	
	Low	20.4 ^a	2.8		10.8 ^a	1.0		20.6 ^a	2.2		5.1 ^a	0.5		4.3 ^a	0.5		3.0 ^a	0.3		2.0 ^a	0.0	
38	Control	25.4 ^b	5.7	0.05	13.8 ^b	1.2	0.05	25.0 ^b	3.1	0.01	6.8 ^b	0.3	0.05	9.2 ^b	0.8	0.05	8.0 ^b	0.8	0.01	5.0 ^b	0.5	0.05
	High	24.9 ^b	5.0		15.0 ^c	1.0		25.9 ^b	2.4		6.0 ^b	0.7		10.1 ^c	1.0		8.5 ^b	1.2		4.7 ^b	0.3	
	Low	20.2 ^a	4.3		10.0 ^a	0.6		19.0 ^a	1.8		4.5 ^a	0.1		7.8 ^a	0.3		6.0 ^a	0.4		4.0 ^a	0.8	
40	Control	26.2 ^b	2.1	0.05	15.0 ^b	1.8	0.05	24.0 ^b	2.3	0.001	5.5 ^c	0.6	0.05	10.4 ^b	0.6	0.05	9.0 ^c	1.0	0.05	5.2 ^c	0.4	0.05
	High	26.0 ^b	3.4		14.5 ^b	1.8		22.8 ^c	1.2		4.1 ^b	0.8		10.9 ^b	1.2		8.0 ^b	1.2		4.0 ^b	0.7	
	Low	20.5 ^a	3.6		11.2 ^a	1.1		16.0 ^a	1.5		3.1 ^a	0.3		8.0 ^a	0.8		7.2 ^a	1.0		3.1 ^a	0.6	

[1] Age (week). [2]M: Means (n=4). [3] SEM: standard error of the mean. [4] |, ^{a-c}: $p < 0.05$ for different letters (trait with treat in different age) according to F Test.

SAS software code for the statistical analysis

Statistic cods:

The normally distributed data were analysed by using *Shapiro-Wilk test*:

Data treatment

Input Body weight;

Cards;

.

.

;

Proc glm;

Class body weight;

Model treatment= body weight/ss3;

Means body weight/hovtest=Bartlett snk;

Output out=residur=residuph P=preditph;

Run;

Quit;

Proc univariate data= residu normal plot;

Var residuph;

Run;

The data of body weight, scrotal circumference were analyzed using the mixed model of SAS 9.4. The effects of treatment duration and treatment-by-time interaction were determined.

data body;

input animal treat\$ weight weight2m age; /* weight 2 month */

cards;

data body2;

set new;

if 2 < age;

run;

proc means data=body2;

run;

proc corr;

run;

proc mixed data=body2;

class animal treat age;

model weight=treat|age weight2m;

random animal;

lsmeans treat|age /pdiff=all;

run;

data Scrotal;

input animal treat\$ scrotum scrotum2m age; /* scrotum 2 month */

cards;

data scrotal2;

set new;

if 2 < age;

run;

proc means data=scrotal2;

run;

proc corr;

run;

proc mixed data=scrotal2;

class animal treat age;

model scrotum=treat|age scrotum2m;

random animal;

lsmeans treat|age /pdiff=all;

run;

Histomorphometrical, spermatogenesis indices and sperm data were analyzed by analysis of variance (ANOVA) using the general linear models (GLM).

data histomorpho;

input animal treatmen\$ age weigh Height width volu len STD GEH SPI RI TDI;

cards;

run;

proc glm;

class treatmen age; where 2 < age;

model weigh Height width volu len STD GEH SPI RI TDI=treatmen|age;

lsmeans treatmen|age /pdiff=all;

run;

Supplementary material to the article “The effect of dietary energy levels on the sexual puberty of ram lambs”, by Farshid Nazari-Zonouz, Gholamali Moghaddam, Gholamreza Hamidian, Hossein Daghigh-Kia and Akbar Taghizadeh. Spanish Journal of Agricultural Research, Vol. 20, No. 3, 2022 (<https://doi.org/10.5424/sjar/2022203-18125>)

```
data Sperm;
input treatmen$ concentration viability volume;
cards;
;
run;
```

```
proc glm;
class treatmen;
model concentration viability volume =treatmen;
lsmeans treatmen /pdiff=all;
run
```

The data of sexual behavior were analyzed using the mixed model of SAS 9.4.

```
Data sexual;
input animal week trt$ weight2m Foot Anog Noising Flehmen Mountat Mounts Mountsej;
cards;
;
run;
```

```
proc means data=sexual;
run;
```

```
proc mixed;
class animal trt week;
model Foot =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model anog =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model Noising =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model Flehmen =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model Mountat =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model Mounts =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```

```
proc mixed;
class animal trt week;
model Mountsej =trt|week weight2m;
random animal;
lsmeans trt|week /pdiff=all;
run;
```