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EFFECT OF DIETARY PROTECTED PROTEIN ON NUTRIENT DIGESTIBILITY AND SOME REPRODUCTIVE PERFORMANCE IN SHEEP.

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SUMMARY

Protein protection methods of canola meal in the present study were done by heat or sodium hydroxide. In the first experiment, the effect of feeding protected protein on digestibility coefficients of different nutrients and nutritive values (Total Digestible Nutrient; TDN, Metabolizable Energy/ ME and Digestible Crude Protein/ DCP) of different tested rations were done using 12 healthy male Sohagi lambs. Animals were divided randomly into three equal treatments (control, heat and sodium hydroxide treatments). Values of TDN, ME and DCP were increased significantly ($P<0.05$) as a result of treatments. In the second experiment, a total number of 36 healthy Sohagi ewes were used. During the experimental period (42 days); animals were divided into three treatments (n=12/ each) as follows: control, heat treatment (T1) and sodium hydroxide treatment (T2). All ewes were mated using fertile ram during breeding season. The values of conception rate, lambing rate, total number of services and number of service per conception were calculated. Blood samples were collected from all ewes in each treatment at estrus during the breeding period to study the effect of protected protein on progesterone concentrations. Results indicated that conception rate was improved by 37.50 % and 25.98 % for T1 and T2, respectively compared with control treatment. The number of service per conception values decreased by 11.04 % and 7.98 % in T1 and T2, respectively compared with control ewes. Lambing rate in control treatment was 112.5%, while this value was improved by 127.27% and 130.00% for T1 and T2, respectively. Serum progesterone concentration was increased in ewes fed ration T1 or T2 (protected protein rations) compared with control ration, but differences were not significant. In the third experiment, a total number of 15 lambs, (age 3-4 months) were used. Animals were divided into the same three (n=5) treatments. Body weight, testes volume and testes circumference were determined at the beginning of the experiment and then month intervals during the experimental period (4 months). Blood samples from each animal in different treatments were collected at the beginning of experiment and then month intervals. Results indicated that testosterone concentrations in the different treatment were increased by 8.86% and 6.89% for treatments T1 and T2, respectively compared with control treatment. Testosterone concentration in ram lambs was increased gradually with advancing age. The highest values of testes circumference and volume recorded in T1 compared with other treatments (T2 and control). Also, both traits were increased gradually with advancing of age. There were significant ($P<0.01$) and positive correlation coefficients among body weight and all traits studied and also, among these traits. Results indicated that protected protein in the diets improved significantly the reproductive performance in male and female sheep.

Keywords: *reproductive performance, protected protein, sheep.*

INTRODUCTION

Sheep flock in Egypt was estimated as 5.4 million heads (Ministry of Agriculture, 2004) which possess a profound economic importance to the agriculture sector. Increasing lambs production can be achieved mainly through increasing the number of lambs per ewe and the number of lambing per year. Generally, as reported in the most literature, insufficient feed intake caused poor growth and decreased reproductive performance of sheep. Gunn *et al.* (1984 a and b) and Abecia *et al.* (1993) found that the ewes fed high level of feeding had higher ovulation rate. There was a general agreement among most authors that nutrition is one of the most important factors affecting the reproductive performance of animals. The effect of feeding levels on prolificacy was primarily due to changes in ovulation rate (Robertson and Hinch, 1990 and Waghorn and Smith, 1990). In addition, Cisse *et al.* (1994) reported that level of feeding found to be an important determinant of goat body condition and correlated positively with the conception rate.

The protein content in diets of ruminant animal is very essential for improving the reproductive performance. Eighty to 90% of the protein content of some feedstuffs with high quality protein may be degraded in the rumen (Beever, 1984), which can be resulted in protein deficiency for maximum reproduction of ruminants (Grigsby *et al.*, 1989). In addition, there is a positive effect of dietary protein on ovulation rate (Abecia *et al.* 1997). Also, El-Reweny (2006) with sheep, El-Sherbieny (2000) and Sayed Ahmed (2000) with Friesian bulls noticed that animals fed protected protein recorded higher testosterone concentrations in blood than animals fed control diet.There were several methods to protect dietary protein from degradation in the rumen. Protein protection methods can be categorized into chemical (e.g. sodium hydroxide, acetic acid and formaldehyde) and physical (e.g. heat) treatments. The effect of heat treatment as a method of protein protection is depending upon both temperature and time of heat exposure. Several authors reported that the benefits of heat treatment are mainly increase rumen undegradable protein in the diets (Griffin *et al* 1992 ; Pires *et al* 1996 and Chouinard *et al*, 1997) . In addition, Stern *et al* (1985) observed an increase in the availability of total essential amino acids in the small intestine of ruminant animals, when the diet contained extruded whole soybeans at 132° C and 149°C .

This investigation was undertaken to study the reproductive performance of Sohagi rams and ewes as affected by protected protein in their ration.

MATERIALS AND METHODS

The present study was carried out at the Experimental Farm of Animal Production Department, Faculty of Agriculture, Sohag University in cooperation with Animal Production Department, Faculty of Agriculture, Minia University. The aim of this study was to investigate the effect of feeding protected protein of canola meal in the rations on reproductive performance of male and female Sohagi sheep as a local breed .

Preparation of experimental rations:

Canola meal is a protein supplement that contains up to 40% crude protein as dry matter basis. Canola meal is one of the best plant protein utilized in feeding either monogastric or ruminants. It presents as well balanced amino aced composition and is widely used in monogastric feeding (Bell, 1984). The only setbacks of canola meal feeding for ruminants would be slight taste aversion observed in some animals and high rate of rumen protein degradability (Krikpatrick and kennelly, 1987). Therefor protein protection in canola meal lades to higher quantity of dietary protein escaping from the rumen fermentation, then increasing the supply of dietary protein reaching the stomach and small intestine and decreased the supply of microbial protein arriving the lower gut of ruminants (Mc Allister et al 1992) Canola meal was used as 25% of the concentrate feed mixture in the present study. The formulation of the experimental concentrate rations are shown in Table (1)

Table (1): Formulation of the experimental concentrate rations.

Item	Treatments		
	Control	T1	T2
Canola meal			
Untreated	25	----	----
Heat treated	----	25	----
Sodium hydroxide treated	----	----	25
Crushed Maize	42	42	42
Wheat bran	30	30	30
Premix*	0.5	0.5	0.5
Sodium chloride	0.5	0.5	0.5
Limestone	2.0	2.0	2.0

** Premix contents per kg are vit. A. 12000000 IU; vit. D3, 2200000 IU; vit. E, 10 gm; vit. K3, 2 gm; copper, 10 gm; zinc, 50 gm; Manganese, 55 gm; Iodine, 1 gm; Selenium, 0.1 gm; Carrier (CaCo3), up to 3000 gm. Control=Canola meal without treatment. T1=Canola meal heat treatment. T2= Canola meal sodium hydroxide treatment*

Methods of protein protection in canola meal:

Protein of canola meal used in the different experimental concentrate rations in the present study was protected by two treatments:-

1- Heat treatment: Two cm layer of ground canola meal was subjected to 135-145° C in a forced air oven for 4 hrs according to Stern *et al* (1985). After heat treatment, canola meal was kept at room temperature (25°C) for 3 days before being and mechanically mixed with other ingredients to formulate the concentrate ration.

2- Sodium hydroxide treatment: Canola meal was spread by solution of sodium hydroxide (10 %) at the rate of 3 gm NaOH/100 gm DM of ground canola meal, according to Mir *et al* (1984). The treated canola meal was air dried at room temperature (25°C) for one week before being and mechanically mixed with other ingredients.

Evaluation of the experimental rations:

In the first experiment, the effect of feeding protected protein on digestibility coefficients of different nutrients and nutritive values (Total Digestible Nutrient/ TDN, Metabolizable Energy/ ME and Digestible Crude Protein/ DCP) of different tested rations, was done using 12 healthy male Sohagi lambs averaged 35.8 ±1.29 kg of body weight digestibility experiment. The experimental period lasted for 3 weeks. Animals were divided randomly into three equal treatments (control, heat and sodium hydroxide treatments). Animals were fed mach concentrate diet (80%) and wheat straw (20%) according to NRC (1985) requirements. Feces samples from each animal were collected at the last week of the experimental period twice daily. Digestibility coefficients of the different nutrients in tested rations were determined using acid insoluble ash (AIA %) as natural marker according to Van Keulen and Young (1977). Total digestible nutrient (TDN) and digestible crude protein (DCP) were calculated.

Female reproductive performances:

In the second experiment, a total number of 36 healthy Sohagi ewes of 3-4 years of age and 40.60± 1.60 kg body weight were used. The ewes were kept in a semi open shed. The animals were divided randomly into three equal (n=12) treatments (control, heat treatment and sodium hydroxide treatment). Ewes were offered their daily requirements from DM , TDN and CP guided by NRC (1985). Ewes in each treatment were group fed during the whole breeding period (42 days). All ewes were mated by one mature fertile ram (about 3 years old) during breeding season. Values of conception rate (number of concepted ewes/ number of ewes exposed x100), lambing rate (number of lambs born/ number of concepted ewes x 100). Total number of service and the number of service per concepted ewes (total number of service / number of concepted ewes) were calculated. Fresh water was available during the experimental breeding period.

Blood samples were collected at estrus day, which was detected by a proven intact mature ram, to study the effect of protected protein on serum progesterone concentrations. Blood samples were allowed to clot at room temperature and serum was then separated by centrifugation at 3000 r.p.m for 15 minutes. A progesterone concentration in serum was performed using active progesterone RIA, DSL-3900 using commercial kits produced by Diagnostic System Laboratories, Inc USA according to Abraham (1981).

Male reproductive performances:

In the third experiment, a total number of 15 male lambs, aged 3-4 month and averaged 16.8- 17.0 ±0.23 kg of body weight were used. Animals were divided randomly into three equal (n=5) treatments (control, heat treatment and sodium hydroxide treatment). Animals were fed during the experimental period on mach concentrate diet (80%) and wheat straw (20%). Daily amounts of DM, TDN and CP offered to animals guided by NRC (1985) . Body weights, testes diameter (testes circumference) and testes volume of lambs were recorded at the beginning of experiment and then at month intervals till the end of experimental period (4 months). Testes diameter was measured by flexible cloth tape at the point of maximum circumference of the paired testes. Testes volume was evaluated from the volume of water they replaced (Salhab *et al.* 2001). Blood samples from each animal in different treatments were collected at the beginning of experiment and then month intervals. Blood samples (about 8 ml / each animal) were collected at the beginning of the experiment and then at month intervals by jugular vein puncture in dry clean glass vials.

Blood samples were allowed to clot at room temperature (25° c) and serum was then separated by centrifugation the samples at 3000 r.p.m for 15 minutes. A total testosterone (T) concentration in serum was performed using active testosterone RIA, DSL- 4000 using commercial kits produced by Diagnostic System Laboratories, Inc USA according to Abraham (1981)

Statistical analysis:

The results were statistically analyzed using the General Linear Model (SAS, 1998) for complete randomized design. Reproductive and blood parameters were performed by methods of analysis of variance. Significant differences among treatments means were analyzed using (Duncan, 1955).

The correlation coefficient between body weight and some reproductive characteristics of male (testes diameter, testes volume and testosterone concentrations) were calculated.

RESULTS AND DISCUSSION

1- Proximate analysis and nutritive values of the experimental ration:

The Proximate analysis of the experimental rations and canola meal are presented in Table (2). Concerning the crude protein (CP) values in different experimental rations (80 % concentrate and 20 % roughage) were 14.69, 14.77 and 14.88 for control, T1 and T2, respectively. Generally, the present results of the proximate analysis of different nutrients in the experimental rations indicated that the protection methods of canola meal led to little differences in the proximate analysis. These results are in agreement with those of Dhiman *et al.* (1997) and El- Reweny (2006). They found that proximate analysis of protected protein in soybean and linseed meals are not affected significantly by different methods applied for protecting protein. Also, the present results indicated that proximate analysis of canola meal used in our study is in agreement with those reported by CLFF (2001).

Table (2): Proximate analysis of the experimental rations and canola meal on dry matter basis.

Item	Treatments			Canola meal
	Control	T1	T2	
OM	91.46	91.46	91.54	92.79
CP	14.69	14.77	14.88	35.62
EE	3.82	3.73	3.73	7.41
CF	13.76	13.14	13.27	12.32
NFE	59.19	59.82	59.66	37.44
Ash	8.54	8.54	8.46	7.21

Control=Canola meal without treatment.
T1=Canola meal heat treatment. T2= Canola meal sodium hydroxide treatment
Rations containing 80% concentrate and 20% roughage.

The primary digestibility trials indicated that the digestibility coefficients of DM, OM, CP, EE and NFE were increased significantly (P<0.05) as a result of protected protein by heat treatment (T1) or sodium hydroxide (T2) in comparison with control treatment, Heat or sodium hydroxide treatments had no effect on CF digestibility coefficients (Table, 3). Generally, improving the digestibility coefficients in T1 and T2 led to a significantly (P<0.05) increase of total digestible nutrient (TDN %), metabolizable energy (ME) and digestible crude protein (DCP %). The improvement of TDN values in T1 and T2 may be due to enhance of digestibility coefficients of CP, EE, CF and NFE in response to the protein protection methods, while the positive effect of protected protein on DCP% may be due to higher digestibility coefficient of CP for these treatments than control. These results are in agreement with those obtained by El-Reweny *(2006)*. He indicated that the nutritive value expressed as TDN% for control ration was significantly lower than treated ration. Also, El-Ayek *et al.* (1999) indicated that the protection of protein source enhanced slightly the digestible crude protein content of the ration of dairy cows from 11.4% for untreated ration to 11.7% for treated formaldehyde ones.

Table (3): Effect of treatments on the nutrients digestibility coefficients and nutritive values of the experimental rations.

Treatments	Digestion coefficients								
	DM	OM	CP	EE	CF	NFE	TDN	ME+	DCP
Control	66.03^{c}	68.04^{c}	67.34^{c}	67.69^{c}	61.23	69.39^{c}	65.27^{b}	2.36^{b}	9.96^{b}
T1	69.77^{a}	71.42^{a}	71.37^{a}	71.30^{a}	61.11	72.51^{a}	68.01^{a}	2.46^{a}	10.62^{a}
T2	67.79^{b}	69.76^{b}	70.31^{b}	69.69^{b}	61.76	71.21^{b}	66.98^{a}	2.42^{a}	10.46^{a}
±SE	1.44*	1.57*	1.19*	1.38*	1.05 NS	1.00*	1.32*	0.01*	0.36*

a, b, c, Means with different letters in the same column are significantly different
** (P< 0.05) NS= Not significant SE = Standard error*
+ME (Metabolizable energy) = TDN X 0.04409X0.82 (Mcal/kg DM)

2- Reproductive performance:

A- Female

The present results indicated that the daily amounts of DM , TDN and CP offered to ewes from different excremental rations ranged between 1.1 – 1.2 k.g , 0.72 – 0.75 Kg and 161- 164 g , respectively .

Reproductive performance of Sohagi ewes fed protected and unprotected protein during breeding period are presented in Table (4). Results indicated that ewes fed rations treated with heat (T1) or sodium hydroxide (T2) showed increase in the conception rates. The values of conception rates were 91.67 % and 83.33 % for T1 and T2, respectively compared with control ration (66.57 %). Thus, conception rate was improved by 137.5 % and 125,00 % for T1 and T2 , respectively compared to control treatment. At the same time heat and sodium hydroxide treatments led to decrease the number of service per conception by 11.04 % and 7.98 % for T1 and T2, respectively compared with control

ewes. The best rate was recorded with T1 (1.45 times) followed by T2 (1.50 times), while the rate recorded for control treatment was 1.63 times (Table, 4). Conception rate improved as a result of feeding protected protein by 37.50% in T1 and 24.98% in T2 in comparison with unprotected dietary protein (control).

Table (4): Effect of experimental diets on some reproductive performance of ewes during the breeding periods.

Item	Treatments		
	Control	T1	T2
No. of ewes	12	12	12
No. of concepted ewes	8	11	10
Conception rate	66.67	91.67	83.33
Total no. of service	13	16	15
No. of services per conception	1.63	1.45	1.50
No. of lambs born	9	14	13
Lambing rate %	112.50	127.27	130.00
Progesterone(P4) (ng/ml) at estrus $^{N.S}$	0.31±0.01	0.33±0.01	0.33±0.01

Control: Canola meal without treatment.

T1: Canola meal heat treatment. T2: Canola meal sodium hydroxide treatment.

Data in Table (4) illustrated that lambing rate were 127.27 % and 130.00 % for T1 and T2, respectively compared with the control 112.50 %. Thus, lambing rate was improved with protected protein treatment. The improvement in the reproductive performance in the present study as a result of protein protection may be due to the positive effect of these treatments on the digestibility coefficients of different nutrients and nutritive values in T1 and T2 as TDN, ME and DCP (Table , 3) . There are positive effects of dietary energy (Smith and Murray, 1995 and Abu El-Ella, 2006) or dietary protein (Abecia *et al.* 1997) on ovulation rate. Moreover, Cisse *et al.* (1994) and Solcuma *et al.* (2006) reported that the level of feeding considered to be an important determinant of goat body condition and correlated positively with the conception rate.

Progesterone concentration increased for ewes feed ration T1 and T2 (protected protein ration) compared with control ration, but the differences were not significant.

B- Male

The presented results indicated that average of daily dry matter, TDN and protein intake values during experimental period in the different treatments (control, T1and T2)

are nearly similar. Daily dry matter intake values during the experimental period in the different experimental treatments were 800.62, 810.72 and 811.87 g/d, and daily TDN intake values were 523, 552 and 544 g/d, while protein intake values from concentrate and roughage were 117.61, 119.74 and 120.81 g/d. for control, T1 and T2, respectively. Differences in dry matter, TDN and protein intake among experimental rations were not significant.

Data of body weight and some reproductive performance (testosterone concentration, testes circumference and volume) of ram lambs during the experimental period (4 months) are presented in Table (5). Values of testosterone concentrations at the beginning and after one month showed insignificant differences among treatments, while after 2, 3 and 4 months there were significant ($P<0.01$) differences. Also results indicated that testosterone level was increased by 8.86% in T1 and 6.89% in T2 compared with control group. These results may be due to increase the nutritive values as TDN, ME and DCP in T1 and T2 treatments as a result of feeding protected protein (Table , 3) . Protected protein led to significantly improvement live body weight (Table, 5) and also, increasing different body organs including the testes, which consider the main source of testosterone secretion in blood. El- Reweny (2006) with sheep noticed that the ram lamb fed protected protein recorded higher (15.3%) testosterone concentrations than lambs fed control diet. Also, El-Sherbieny (2000) and Sayed- Ahmed (2000) found that Friesian bulls fed diets contain protected protein showed a higher and significant ($P<0.05$) testosterone concentration in their blood plasma than that of the control.

Table (5): Effect of protected protein methods on means of body weight and some reproductive performance of ram lambs during the experimental period.

Item	Treatments (LSM)			±SE
	Control	T1	T2	
Body weight (kg)	25.90 ^{c}	28.62 ^{a}	27.44 ^{b}	0.28**
Testosterone(ng/ml)	2.03 ^{b}	2.21 ^{a}	2.17 ^{a}	0.03**
Testes circumference(cm)	19.02 ^{c}	21.06 ^{a}	20.17 ^{b}	0.28**
Testes volume(ml)	118.44 ^{c}	148.68 ^{a}	131.36 ^{b}	4.63**

*a, b, c Means with the different letters in same row are significantly different *(P<0.05), **(P<0.01), NS = Not significant. SE = Standard error*

In the present study, testosterone hormone level of ram lambs was increased gradually with advancing age (Table 6). These results may be due to the increment of body weight and testis weight as a result of age progress. These results are in agreement with those of Miller *et al.* (1989) who found that serum testosterone levels were increased gradually in ram lambs during the period from 24 to 30 weeks of age.

They indicated also that the average of testosterone concentration at 12, 18, 24 and 30 weeks of age were 1.46, 2.56, 5.32 and 7.44 (ng/ml) in blood serum, respectively.

Table (6): Effect of age on body weight and some reproductive performance of ram lambs during experimental period.

Item	Body weight (kg)	Testosterone (ng/ml)	Testes circumference (cm)	Testes volume (ml)
At the beginning	17.43 ^{e}	0.79 ^{d}	12.47 ^{e}	51.73 ^{e}
One month	22.17 ^{d}	2.05 ^{c}	16.74 ^{d}	95.33 ^{d}
2 months	28.67 ^{c}	2.35 ^{c}	22.06 ^{c}	142.33 ^{c}
3 months	31.83 ^{b}	2.62 ^{b}	23.64 ^{b}	177.80 ^{b}
4 months	37.10 ^{a}	2.88 ^{a}	25.50 ^{a}	196.93 ^{a}
±SE	±0.36**	±0.04**	±0.63**	±6.55 **

*a, b, c, d Means with different letters in the same column in each parameter are significantly different, ** (P< 0.01).*

Also, Salem (1997) reported that testosterone levels in sheep increased in amplitude and frequency with advancing age until reached 3.48 ng/ml at 375 days of age (puberty). Such age effect on plasma testosterone was reported by Miyamoto *et al.* (1989); Mokhless and Ibrahim (1990) and Abu-Elawa (1995) with cattle and buffalo and El-Reweny (2006) with sheep. They reported that the rise of plasma testosterone concentration with advancing of age due to the development of the testis as a result of age progress.

Concerning the effect of protected protein methods on testes circumference and testes volume, the results in Table (5) indicated that T1 and T2 recorded higher values of both diameters compared with control group. These results may be due to improving TDN, ME and DCP in T1 and T2 (Table, 3) led to the increase of live body weight and both testes diameters. Also, several investigators found significant relationship between body weight and testicular diameters (Abu-Elawa, 1995 and Bayoumi, 1999 with buffalo and Friesian calves and Abd El-Hakeam, 1978 with sheep). The present results are in agreement with those of El-Ayek *et al.* (1999). They reported that lambs fed protected protein had higher testicular volume, testes weight and testo-somatic index than control. Also, El-Reweny (2006) observed that the scrotal circumference of lambs fed protected protein increased significantly in comparison with lambs fed control diet.

In the present study, increase of testosterone concentrations, diameter and volume of ram lambs testis gradually with advancing age (Table, 6) may be due to the increase in body weight and testicular measurements development with advancing of age. These results are in agreement with those of Muskasa and Ezaz (1992). They reported that scrotal circumference of ram was increased gradually and correlated with age and body weight. Also, Salem (1997) observed that circumference in Saidi ram lambs increased gradually with advancing of age from 1-2 months old until reached puberty.

In addition, Salhab *et al.* (2001) added that age and live body weight of Awassi ram lambs were positively correlated with testicular circumference and testicular volume. The same results were obtained by Hamdon (2005).

The correlation coefficients between body weight and each of testes circumference, testes volume and testosterone level in one hand and on the other hand among all the reproductive traits are highly (P<0.01) significant (Table 7). These results revealed that the developments of evacuation organs, age and testes size were directly correlated with the testosterone section coming from the testis. These results are in agreement with Abd El-Hakeam (1978); Salhab *et al.* (2001), Ozturk *et al.* (2002) and Hamdon (2005). They found that the correlation between age, live weight and testicular measurements in sheep were statistically significant (P<0.01). Similar results were obtained by Abu-Elawa (1995) in both buffalo and cattle.

Table (7): Correlation coefficients between body weight and some reproductive performance of male during experimental periods.

Measurements	Testes circumference	Testes volume	Testosterone
Body weight	0.936**	0.950**	0.912**
Testes circumference	-	0.966**	0.884**
Testes volume	-	-	0.868**

*** Significant, (P< 0.01). n = 75*

Generally, from the present result it can be concluded that the protected protein significantly improved the reproductive performance in the male and female of sheep.

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تأثير البروتين المحمي في العلائق على هضم المركبات الغذائية و بعض المظاهر التناسلية في الأغنام

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تم في الدراسة الحالية حماية البروتين لكسب الكانولا باستخدام الحرارة أو الصودا الكاوية .

التجربة الأولى تم تقييم تأثير حماية البروتين على معاملات هضم المركبات الغذائية و القيمة الغذائية (TDN, ME , DCP) للعلائق المختلفة وقد تم عمل تجربة هضم باستخدام 12 ذكر من اغنام السوهاجي وقد تم تقسيم الحيوانات الى ثلاثة مجموعات متساوية وقد اظهرت النتائج زيادة معنوية في كل من TDN, ME , DCP نتيجة المعاملة ؛ التجربة الثانية أجريت باستخدام عدد 36 من النعاج قسمت خلال فترة التجربة (42يوم) إلى ثلاثة مجموعات متساوية هي : الكنترول – الحرارة (المعاملة الأولى)–الصودا الكاوية (المعاملة الثانية) . وقد تم تغطية الاحتياجات الغذائية للنعاج باستخدام العليقة المركزة وقش الأرز طبقاً لمقررات NRC (1985) . لقحت النعاج باستخدام كبش مختبر مخصب وذلك خلال موسم التناسل وتم حساب كل من معدل الإخصاب ومعدل الولادات والعدد الكلي للتلقيحات وكذلك عدد التلقيحات اللازمة لكل حمل مخصب كما تم أخذ عينات من الدم ثناء دورة الشبق لقياس تركيز هرمون البروجستيرون . وقد أظهرت النتائج أن نسبة الإخصاب تحسنت بنسبة 37.5 % و 25.98 % في المعاملة الأولى والثانية مقارنة بمعاملة الكنترول . كما أن عدد التلقيحات اللازمة لحدوث الحمل المخصب كانت قليلة في تلك المعاملات بالمقارنة بالكنترول . معدل الولادة في المعاملة الكنترول كان 112.50 % بينما معدل الولادة تحسن بنسبة 127.27 % ، 130.0% في المعاملة الأولى والثانية. حدثت زيادة في تركيز هرمون البروجستيرون في المعاملة الأولى والثانية مقارنة بالكنترول ولكن الفروق كانت غير معنوية .

التجربة الثالثة أجريت باستخدام عدد 15 حمل ذكر عمر 3-4 شهور . قسمت الحملان إلى ثلاث مجموعات وسجلت قيم وزن الجسم وحجم الخصية ومحيط الخصية في بداية التجربة ثم شهرياً حتى نهاية فترة التجربة بعد 4 شهور . أخذت عينات من الدم من الحملان في بداية التجربة ثم شهرياً لتقدير تركيز هرمون التيستيرون في الدم . أظهرت النتائج أن تركيز الهرمون يزداد بمعدل 8.86% ، 6.89% في المعاملة الأولى والثانية مقارنة بالكنترول خلال فترة التجربة ككل . كما أن مستوى التيستيرون في الدم يزداد بزيادة العمر . و سجلت القيم الخصة بحجم ومحيط الخصية قيم مرتفعة في المعاملة الأولى مقارنة بالمعاملة الثانية والكنترول . وقد حدثت زيادة أيضا في كلا المقياسين بزيادة العمر . وجد أن هناك ارتباط معنوي موجب بين وزن الجسم وجميع المقاييس المسجلة على الذكور كما وجد أيضاً أن هناك ارتباط بين تلك المقاييس وبعضها .

وقد أظهرت نتائج هذه الدراسة أن استخدام البروتين المحمي يؤدي إلى تحسين معنوي للخصائص التناسلية في إناث وذكور الأغنام .