

Influence of Varying Levels of Dietary Undegraded Intake Protein Intake on Nutrient Intake, Body Weight Change and Reproductive Parameters in Postpartum Awassi Ewes

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ABSTRACT : The objective of this study was to evaluate the effect of dietary undegradable protein (UP) level on body weight change, nutrient intake, milk production and postpartum reproductive performance of Awassi ewes. Twenty-seven multiparous Awassi ewes (initial body weight = 53.3±1.6 kg) were randomly assigned to three dietary treatments (9 ewes/treatment) for 62 days using a completely randomized design. Experimental diets were isonitrogenous (15.5% CP), isocaloric, and were formulated to contain 17.9 (LUP), 27.1 (MUP), and 34.0% (HUP) of the dietary CP as UP. On day 10±3 (day 0 = parturition) ewes were housed in individual pens for 5 weeks. Feed offered and refused was recorded daily. At the end of this period, animals were removed from their pens and combined into 3 separate groups (LUP, MUP and HUP). One fertile, harnessed ram was allowed with each group for 34 days. Rams were rotated every 2 days among the three groups. Each group was offered the corresponding experimental diet. Organic matter, CP, UP and metabolizable energy intakes were higher ($p<0.05$) for ewes fed the HUP diet compared with ewes fed the LUP and MUP diets. Ewes fed the HUP diet gained more ($p<0.05$) weight compared with ewes fed the MUP diet (7.3 vs. 2.1 kg), while ewes fed the LUP diet lost an average of 2.1 kg. Pregnancy rate of ewes fed the HUP diet was 100%, compared with 66 and 33% for ewes fed the MUP and LUP diets, respectively. Lambing rate was greater ($p<0.05$) for ewes fed HUP (8/9) diet compared with ewes fed the MUP (4/9) and LUP (3/9) diets. These results indicate that Awassi ewes receiving adequate dietary UP level consume more feed and are capable of returning to estrus shortly after parturition and are capable of producing two lamb crops per year. (*Asian-Aust. J. Anim. Sci.* 2005, Vol 18, No. 5 : 637-642)

Key Words : Bypass Protein, Awassi, Sheep, Reproduction, Undegradable Protein

INTRODUCTION

Awassi is the most numerous and widespread sheep breed in West Asia. Awassi is the predominant breed in Iraq (Al-Haboby et al., 1999), the most important in Syria (Zarkawi, 1997), Turkey (Vanli and Ozsoy, 1989) and the only indigenous sheep breed in Lebanon (Hamadeh et al., 2001) and Jordan (Zarkawi, 1997). Available data from this region on Awassi sheep breeding season is highly variable. Some of this variability could be attributed to the nutritional status of the animal (Kridli et al., 2001).

Both energy (Senator et al., 1996) and protein quantity and quality (Wiley et al., 1991) have been shown to influence postpartum (PP) reproductive performance. Canfield and Butler (1989) reported that energy balance played an important role in the re-establishment of ovarian activity in dairy cows. Dhuyvetter et al. (1993) concluded that beef cattle fed 50% of the crude protein (CP) requirements as undegradable intake protein (UP) lost less body weight than cows fed 25% of the CP as UP, and the interval from calving to first estrus was reduced. Kridli et al. (2001) fed PP Awassi ewes 2 diets containing 20 and 35%

of the dietary CP as UP and observe that ewes fed the 35% UP diet gained more weight and showed more luteal activity compared with ewes fed the 20% UP diet. Our objective was to evaluate the effect of dietary UP content on body weight change, nutrient intake, milk production and postpartum reproductive performance of Awassi ewes.

MATERIALS AND METHODS

This experiment was conducted at Jordan University of Science and Technology during the months of December and January 2002/2003. Average day length in December and January is 10 h 29 min and 11 h 12 min, respectively. The average minimum and maximum ambient temperatures for December and January are 10°C and 17°C and 4°C and 12°C, respectively.

Twenty-seven multiparous, winter-lambing Awassi ewes (initial body weight = 53.3±1.6 kg) and their lambs were randomly allotted to one of three dietary treatments in a completely randomized design (9 ewes/treatment). Ewes were assigned to treatments 10±3 days PP (day 0 = parturition). All ewes were nursing single lambs. The experiment was divided into two periods; feeding and mating.

Feeding period

The feeding period lasted for 4 weeks during which

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Table 1. Ingredient composition of the experimental diets

Ingredient (% of DM)	Diet*		
	LUP	MUP	HUP
Corn	4	19.5	25.5
Barley	37	17	4
Wheat bran	17	7	4
Soybean meal	0	9.5	17.5
Wheat straw	37	43	45.5
Urea	2	1.3	0.5
Mineral and vitamin mix ¹	2	1.9	2
Dicalcium phosphate	0.5	0.4	0.5
Salt	0.5	0.4	0.5

* Contained 17.9, 27.1 and 34% of the dietary CP as UP in the LUP, MUP and HUP diets, respectively.

¹Supplies per kilogram of feed: 4.9 mg of Zn, 4.05 mg of Mn, 0.45 mg of Cu, 0.075 mg of I, 0.1 mg of Se, 2,500 IU Vitamin A, 400 mg of Vitamin D, 2.5 IU Vitamin E.

ewes and their lambs were placed in individual pens (1.5×0.75 m²) in early December and fed the experimental diets twice daily. Animals were allowed one week in the individual pens for adjustment before receiving the experimental diets.

The UP contents of the experimental diets were low (LUP), medium (MUP) and high (HUP). The LUP, MUP and HUP diets contained 17.9, 27.1 and 34.0% of the dietary CP as UP, respectively. Soybean meal was used to increase the UP content in the MUP and HUP diets (Table 1). The three experimental diets were isonitrogenous, isocaloric (Table 2). Diets were fed *ad libitum*.

Experimental diets were mixed for the whole period and sampled at the time of mixing. Amounts of feed offered and refused were recorded daily. Feed offered was not accessible for lambs. Clean drinking water was available in plastic buckets. Animal pens were cleaned weekly. Body weights of ewes and lambs were recorded weekly. Animals were maintained at ambient temperature and natural day length throughout the experiment.

Blood samples were collected from each ewe via jugular venipuncture into heparinized tubes (5 IU heparin/tube) every other day during this period to monitor progesterone profiles. Blood was allowed to clot at room temperature. Serum was harvested by centrifugation and stored at -20°C until analyzed for progesterone by radioimmunoassay (Diagnostic Products, Los Angeles, USA). Progesterone samples were run in a single assay. Intra-assay CV was 4.9%, and the sensitivity of the P₄ assay was 0.1 ng/ml. The first observed elevation in progesterone above 1.0 ng/ml constituted the "progesterone rise" which is an indicator of luteal activity. Pregnancy was determined based on sustained progesterone elevation above 3.0 ng/ml on days 18 to 21 post estrus.

Composite samples of diets were oven-dried (60°C), ground through 1 mm screen and analyzed for CP (AOAC, 1990), neutral detergent fiber (NDF) and acid detergent fiber (ADF) (Goering and Van Soest, 1970). The dietary UP

Table 2. Chemical composition of the experimental diets

Item	Diet*		
	LUP	MUP	HUP
DM (%)	93.6	93.1	93.0
Organic matter (% of DM)	89.4	89.9	89.9
Crude protein (% of DM)	15.5	15.5	15.6
UP ¹ (% of dietary CP)	17.9	27.1	34.0
NDF (% of DM)	43.5	43.1	42.9
ADF (% of DM)	22.9	24.9	25.8
Metabolizable energy ² (Mcal/kg)	2.26	2.26	2.26

* Contained 17.9, 27.1 and 34% of the dietary CP as UP in the LUP, MUP and HUP diets, respectively.

¹UP = undegradable intake protein: calculated using NRC (2000).

²Calculated using NRC (1985).

(% of CP) estimates for corn, barley, bran, and soybean meal were 55, 27, 20 and 34%, respectively (NRC, 2000). Metabolizable energy content of the diets was estimated using NRC (1985).

Mating period

At the end of the individual feeding period, animals were removed from their pens, combined into three separate groups (LUP, MUP and HUP) and were offered the same corresponding diet *ad libitum*. One fertile harnessed ram with color marker was allowed with each group for 34 days. Rams were rotated every 2 days among the three groups. Blood samples were collected every third day to monitor progesterone profile. Ewes were checked for breeding marks on a daily basis during this period. Lambing data were recorded 6 months later.

Estimation of milk production

Milk production was estimated in all ewes by oxytocin-induced hand milking (Said et al., 1999) on day 45 (end of feeding period) and day 79 (end of mating period) postpartum. On the days of milking, each ewe was injected intravenously with oxytocin (5 IU) and then directly milked by hand. Ewes were isolated from their lambs for four hours after which the milking procedure was repeated. At this time, milk production was evaluated using a graduated cylinder. A 24-hour milk production was estimated based on this data.

Statistical analysis

Means for diet effects were analyzed as a completely randomized design using the general linear model procedure of SAS (SAS, 1991). Differences among treatment means for the experiment were detected by least significant difference (SAS, 1991). The effect of treatment on the expression of estrus and pregnancy were analyzed by χ^2 -test.

RESULTS

The LUP, MUP and HUP diets contained 0, 9.5 and

Table 3. Body weight changes and nutrient intake of postpartum Awassi ewes fed diets with variable protein degradabilities

Item	Diet*			SE
	LUP	MUP	HUP	
DM intake (g/d)	1,652 ^c	2,105 ^b	2,511 ^a	126
OM intake (g/d)	1,545 ^c	1,893 ^b	2,293 ^a	123
CP intake (g/d)	256 ^c	326 ^b	392 ^a	43
UP ¹ intake (g/d)	49 ^c	88 ^b	133 ^a	19
NDF intake (g/d)	719 ^c	907 ^b	1078 ^a	109
ME ² intake (Mcal/d)	3.74 ^c	4.76 ^b	5.68 ^a	0.6
Initial weight (kg)	53.2	52.7	53.9	1.8
Final weight (kg)	51.1 ^a	55.1 ^a	61.2 ^b	2.0
Body weight change (kg)	-2.1 ^c	2.4 ^b	7.3 ^a	1.4
Milk production (l/d)				
45 d postpartum	1.4	1.2	1.6	0.1
79 d postpartum	1.4	1.1	1.5	0.1

* Contained 17.9, 27.1 and 34% of the dietary CP as UP in the LUP, MUP and HUP diets, respectively.

¹UP = undegradable intake protein.

²ME = metabolizable energy.

^{a,b,c} Means within the same row with different superscripts differ ($p < 0.05$).

17.5% soybean meal, respectively. The chemical composition of the experimental diets is shown in Table 2. Crude protein content was similar for the three diets (15.5, 15.5 and 15.6% for LUP, MUP and HUP diets, respectively). Metabolizable energy content was similar among the experimental diets (2.26 Mcal/kg DM).

Dry matter intake was higher ($p < 0.05$) for ewes fed the HUP diet (2,511 g/d), compared with ewes fed the MUP (2,105 g/d) and LUP (1,652 g/d) diets. Similar results were observed for OM, NDF and metabolizable energy intakes (Table 3). Crude protein intake was higher ($p < 0.05$) for ewes fed the HUP, compared with ewes fed the MUP and LUP diets. The UP intake was higher ($p < 0.05$) for ewes fed the HUP (133 g/d) as compared to ewes fed the MUP (88 g/d) and LUP (47 g/d) diets. Initial body weights of ewes were similar (Table 3). Ewes fed the HUP diet gained more weight ($p < 0.05$) compared with ewes fed the MUP diet (7.3 vs. 2.4 kg), while ewes fed the LUP diet lost an average of 2.1 kg.

Serum samples collected every other day were analyzed for progesterone concentration. Concentrations above 1.0 ng/ml were considered to be indicative of luteal activity.

There was no significant ($p > 0.05$) difference among all dietary treatments on the day PP to first progesterone rise, and days PP to ovulation (Table 4). The first PP estrus was not detected as ewes were still in the individual pens (feeding period) at the time of the first rise in plasma progesterone. Observed estrus (second estrus) was detected at a similar time for all dietary treatments (53.3, 52.1 and 57.9 days PP for ewes fed the LUP, MUP and HUP diets, respectively). The number of ewes expressing estrus was similar for the three treatments (Table 4). Ewes fed the HUP diet had greater ($p < 0.05$) pregnancy rate compared with the ewes fed the LUP diets (9/9 and 3/9, respectively). Lambing rate was greater ($p < 0.05$) in ewes fed the HUP diet (8/9) compared with ewes fed the MUP and LUP diets (4/9 and 3/9, respectively) while the number of lambs born per ewe was similar among treatments (Table 4).

DISCUSSION

The breeding season for Awassi sheep in Jordan begins in June and lasts until September peaking in July (Epstein, 1982). Lambing season begins in late October and peaks in December. Moreover, grass growth and optimum pasture availability occur in February and March. Thus, peak lambing does not coincide with the availability of the better nutrition. Therefore, lactating ewes are faced with inadequate nutrition, loss of body weight and undergo lactational and nutritional anestrus. When ewes graze the spring pasture, their body condition is restored allowing them to start cycling in May/June (Kridli et al., 2001).

Supplying PP ewes with a high level of UP in the early PP period allowed them to consume more feed and gain more body weight (Kridli et al., 2001). In the current study, DM and UP intake for ewes fed the HUP diet were higher compared with ewes fed the LUP and MUP diets. It is clear that these higher intake levels observed with the HUP diet contributed to the increased body weight of ewes fed this diet compared with MUP. This is consistent with findings of other researchers who reported high weight gains in ewes (Kridli et al., 2001) and beef cattle (Wiley et al., 1991) supplemented with UP from various sources. Similarly,

Table 4. Postpartum reproductive performance of Awassi ewes fed diets with variable protein degradabilities

Item	Diet*		
	LUP (n = 9)	MUP (n = 9)	HUP (n = 9)
Number of ewes expressing estrus	7	8	9
Days postpartum to ovulation** (mean±SE)	43.7±2.5	39.5±2.5	40.8±2.5
Days postpartum to first P4 rise (mean±SE)	47.7±2.5	43.5±2.5	44.8±2.5
Pregnant ewes	3 ^b	6 ^{ab}	9 ^a
Lambing ewes	3 ^b	4 ^b	9 ^a
Number of lambs per exposed ewe (mean±SE)	0.3±0.16 ^b	0.4±0.16 ^b	1.0±0.16 ^a

* Contained 17.9, 27.1 and 34% of the dietary CP as UP in the LUP, MUP and HUP diets, respectively.

** Calculated based on progesterone profiles.

^{a,b} Means within the same row with different subscripts differ ($p < 0.01$).

Hoaglund et al. (1992) reported that improvement in body weight and nutritional status of ewes fed soybean meal compared with those fed urea was probably related to either enhancement of microbial activity and (or) increased quantity of protein reaching the small intestines. Enhancement of microbial activity may result from providing growth limiting organic and amino acids for the microbial population in the rumen, which may improve microbial yield.

It is not likely that the difference in DMI was due to the difference in starch source in the experimental diets. Hadjipanayiotou (2004) concluded that mixtures of corn or barley grain attain similar DMI and similar milk yields, although high barley diets may result in some digestive upsets. However, the high urea content of the LUP diet may have contributed to the lower DMI observed for ewes consuming this diet.

Postpartum weight change is more important to reproductive success than the absolute body condition of the animal (Randel, 1990). Oqla et al. (2004) concluded that maintaining body weight in postpartum Awassi ewes allowed the resumption of reproductive activity. Similarly, Rutter and Randel (1984) reported that cows maintaining their body condition during the PP period had shorter PP interval. In the current study, body weight change did not affect the PP interval. Canfield and Butler (1989) reported that energy balance was important for re-establishment of ovarian cyclicity in dairy cows by decreasing LH pulse frequency until the animal returned toward positive energy balance. This is consistent with other researchers who reported that underfed PP beef cows had a loss in body weight that was related to the lack of ovarian activity which appears to be due to the suppression of LH pulse frequency from anterior pituitary gland, which in turn is controlled by the release of the GnRH from hypothalamus (Richards et al., 1989; Randel, 1990). In the current study, ewes fed the HUP diet gained more weight and had higher lambing rate compared with the ewes fed MUP and LUP diets. Ruminally undegradable protein is one of the limiting factors affecting milk yield and ovarian functions during early lactation of dairy cows (Kanjanapruthipong and Buatong, 2002). Al-Haboby et al. (1999) reported similar result for grazing Awassi ewes. Ruminally undegradable protein may improve productivity through altering the metabolizable protein: metabolizable energy ratio (Chowdhury et al., 2002).

Detrimental effects of under-nutrition on reproduction in female animals could be exerted at the ovarian level, anterior pituitary and/or hypothalamus (Schillo, 1992). Under-nutrition during the PP period causes anestrus through several possible mechanisms. These include impaired ovarian response to LH (Canfield et al., 1990).

reduced pituitary storage, synthesis, and secretion of LH (Landfield et al., 1989), reduced pituitary responsiveness to GnRH (Kile et al., 1991), and reduced pulsatile release of GnRH (Miller et al., 1998). Gonadotropin releasing hormone-induced LH release is dependent upon the availability of energy (Sen et al., 1979).

In the current study, losing body weight may have decreased nutrient availability required to influence activity of neurons that control GnRH release. Nutrient availability of metabolic fuels such as glucose (Bucholtz et al., 1996), insulin (Harrison and Randel, 1986), free fatty acids (Estienne et al., 1990) and amino acids (Zurek et al., 1995) may relay messages about the nutritional status of the animal to the GnRH-producing cell. Miller et al. (1998) concluded that nutritional stimulation of gonadotropin secretion was accompanied primarily by fluctuation in plasma and cerebrospinal fluid concentrations of insulin, glucose, free fatty acids and certain amino acids in adult sheep. These correlated changes indicate that the brain may monitor metabolic fluctuation and are part of the critical link between the control system regulating metabolic status and the reproductive axis. Schillo (1992) reported that pulsatile GnRH release is regulated by specific metabolites and (or) metabolic hormones that reflect the nutritional status of the animal, and availability of oxidized metabolic fuels may influence the activity of neurons that control GnRH release.

As stated earlier, ewes fed the HUP diet had a higher pregnancy rate, but not time to first ovulation or oestrus. Clearly, there has been a failure of fertilization; implantation or early embryonic survival for ewes fed the LUP and MUP diets. A possible explanation for this is the higher non-protein nitrogen content of these diets which is usually accompanied with higher blood ammonia levels. Bishonga et al. (1994) concluded that ammonia toxicity for embryos in cows was caused by feeding high levels of rumen degradable protein. Similar results were reported for cows (Butler et al., 1996).

In conclusion, this study demonstrates that feeding diets containing higher amounts of soybean meal as a source of UP has a positive effect on body weight and fertility in PP Awassi ewes. Awassi ewes are capable of cycling during the early PP period provided that adequate nutrition is available to minimize loss in body weight. Feeding diets that contain high levels of UP improves both pregnancy and lambing of Awassi ewes.

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