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REGULAR ARTICLES

Effects of supplemental lysine and methionine on performance of nursing Awassi ewes fed two levels of dietary protein

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Abstract

The objective of this study was to investigate the effects of rumen-protected lysine (RPL) and methionine (RPM) supplementation on production performance of nursing ewes fed two levels of dietary protein. Individually housed Awassi ewes ($n = 34$) nursing single lambs were randomly assigned (2×2 factorial design) to one of four dietary treatments with two levels of protein (170 or 151 g/kg; HP or MP) and two levels of RPL and RPM (0 or 8.5 plus 4 g/day/ewe of RPL and RPM, respectively; no or yes). The trial lasted for 5 weeks. Ewes fed the MP diets had ($P < 0.01$) lower protein intake compared to those fed the HP diets. Intake of other nutrients and milk composition were not significantly ($P > 0.13$) affected by dietary treatments. Ewes fed the MPYES diet produced more ($P < 0.05$) milk compared to those fed the MPNO and HPYES diets and tended ($P = 0.08$) to be more than the HPNO diet. Additionally, milk composition yields for the MPYES diet were significantly ($P < 0.05$) more than the HPYES diets and tended ($P \leq 0.10$) to be more than the MPNO and HPNO diets. Milk efficiency was highest ($P < 0.05$) for the MPYES diet. Final BW, total gain, and growth rate of lambs were greater when their dams were fed the MPYES diet compared to MPNO and HPNO diets. Under our conditions, decreasing dietary protein from 170 to 151 g/kg did not negatively affect the performance of ewes and their lambs. Supplemental RPL and RPM were beneficial for ewes fed diets containing 151 g/kg, but not 170, protein.

Keywords Nursing sheep · Dietary protein · Lysine · Methionine

Abbreviations

AA	Amino acid
ADG	Average daily gain
ARTD	Agricultural Research and Training Department
BW	Body weight
CP	Crude protein
DMI	Dry matter intake
HP	High protein
JUST	Jordan University of Science and Technology
L	Lysine
ME	Metabolizable energy
M	Methionine
MP	Moderate protein
RPL	Rumen-protected lysine
RPM	Rumen-protected methionine

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Introduction

Worldwide, there has been great interest among ruminant nutritionists to decrease dietary protein contents without negatively impacting the production levels of animals (Bahrami-yekdangi et al., 2016). This should not only decrease production cost, but also decrease nitrogen (ammonia) emission into the environment. One of the most appropriate approaches to achieve this goal is by applying amino acid (AA), rather than protein, balancing (Schwab and Broderick, 2017). Simply decreasing dietary protein without balancing AA in metabolizable protein supply will decrease animal production which is disadvantageous.

Amino acid balancing is based on meeting AA requirements, and not necessarily protein per se, for optimal animal performance. Amino acid balancing offers several benefits (Patton et al., 2014; Abbasi et al., 2018, 2019) as it can decrease production cost (via decreasing protein contents of the diet), environmental pollution (via decreasing urea excretion), and animal energy-expenditure (via decreasing urea synthesis). Ruminants utilize dietary protein with different efficiencies based on many factors including dietary protein content, level of individual AAs, and level of limiting AAs (NRC, 2001). Lysine (L) and methionine (M) are the most common AAs that limit milk and milk protein production in dairy cows (and sheep) because they are the most likely AAs to be deficient in intestinally available protein in most dairy diets (NRC, 2001; Park et al., 2020; Kim and Lee, 2021). Up to date, AA requirements for small ruminants have not been determined (Tsiplakou et al. 2020).

To apply AA balancing in ruminants, there is a need to use ruminally protected AAs such as ruminally protected L (RPL) and M (RPM). Using protected AA is a must as unprotected AAs are readily degraded by ruminal microbes. The use of RPL and/or RPM in dairy cows has been extensively investigated and provided variable improvements in production, health, and fertility (NRC, 2001; Patton, 2010; Kim and Lee, 2021). Although it is inconsistent, RPL and/or RPM improved milk production, composition (e.g., protein and fat contents), and composition yield of dairy cows (Robinson, 2010; Awawdeh, 2016; Schwab and Broderick, 2017). A few reports have demonstrated that RPM improved milk yield in dairy ewes (Goulas et al., 2003) and goats (Flores et al., 2009; Titi, 2017). Recently, supplying Awassi ewes with RPL and RPM during the flushing period improved their reproductive performance (Kasim et al., 2020). Data on RPL and RPM, particularly with low-protein diets, in dairy sheep is scarce (Ayyat et al., 2021).

With AA balancing, it was feasible to decrease dietary protein content (from 180 to 160 g/kg and from 164 to 148 g/kg) without negatively affecting milk production of dairy cows (Bahrami-yekdangi et al., 2016). It has been demonstrated that feeding dairy cows diets deficient in metabolizable protein, but supplemented with RPL and RPM, did not depress milk yield (Lee et al., 2012). Supplemental RPL and RPM improved milk yield and composition (protein, fat, and lactose contents) of Comisana ewes fed diets with two levels (157 or 129 g/kg) of dietary protein (Sevi et al., 1998).

In light of this background, we hypothesize that dietary protein levels can be reduced when RPL and RPM are supplemented to lactating ewes without negatively impacting their productive performance. The objective of this study was to investigate the effects of supplemental RPL and RPM on milk yield and composition of nursing Awassi ewes fed two levels of dietary protein and on their lamb growth rate.

Materials and methods

The Institutional Animal Care and Use Committee at Jordan University of Science and Technology (JUST) approved all procedures used in this study (#16/3/2/680). The study was conducted at the Animal Field (Agricultural Research and Training Department-ARTD; semi-arid area, 32° 30′ N, 35° 57′ E, and 510 m above sea level) at JUST.

Animals and design

Thirty-four Awassi ewes (initial BW = 58.1 ± 5.6 kg, age = 4.0 ± 1.5 years, parity = 3.0 ± 1.0; average ± SD) nursing single lamb were utilized in this study. The average lambing date for all ewes was 10 days before starting the study. Ewes and their lambs were individually housed in shaded pens (1.5 m × 0.75 m). Ewes and their lambs were randomly assigned to one of four dietary treatments according to the 2 × 2 factorial design with two levels of protein (170 or 151 g/kg; HP or MP) and two levels of RPL plus RPM (0 or 8.5 plus 4 g/day/ewe of RPL and RPM, respectively; no or yes). Initial BW and milk yield of ewes were considered in randomization.

Diets were formulated to be isocaloric and to meet all nutrient requirements for nursing ewes (NRC, 2007). Ewes were fed their corresponding diet as total mixed ration (TMR) twice a day (equal proportions at 08:00 and 15:00 h) and had free access to clean water throughout the study. Supplemental RPL and RPM were top-dressed daily on offered morning feed to ensure complete consumption. Based on manufacturers' guidelines, the offered amount of RPL (8.5 g/day) and RPM (4 g/day) provided 1.7 and 1.0 g/day of metabolizable L and M, respectively. The amount of offered RPL and RPM was determined to achieve a ratio of 3:1 (L to M) in metabolizable protein for optimal milk protein synthesis (NRC, 2001). The amounts of offered and refused feed were recorded daily for each pen and were adjusted to ensure ad libitum consumption (refusal of about 10% of intake). Lambs had limited access to their dams' feed, and thus, their performance was not exclusively dependent on their dams' milk.

The study lasted for 5 weeks including 1-week adaptation to treatment diets. Ewes and their lambs were weighed before the morning feeding at the beginning and weekly throughout the study. Ewe body weight change and lamb growth rate were estimated accordingly. Milk yield of ewes was recorded weekly throughout the study using double oxytocin (10 IU) injection (IM) and hand milking as described previously (Said et al. 1999). Individual milk weights were recorded, and 125 ml sample was collected and immediately analyzed for composition.

Sample collection and analytical methods

For each pen, samples of refused feed were collected daily, composited at the end of study, and saved for later analysis (AOAC, 1990 methods) for dry matter (DM; method 967.03), organic matter (OM; method 942.05), N (Kjeldahl procedure; method 976.06), and ether extract (EE; method 920.29). Samples were also analyzed (ANKOM^{2000} fiber analyzer, ANKOM Technology Corporation, Fairport, NY, USA) for neutral detergent fiber (NDF; with heat stable α-amylase and sodium sulfite) and acid detergent fiber (ADF) according to Van Soest et al. (1991), and values were expressed inclusive of residual ash.

Milk samples were analyzed for composition (SNF, lactose, crude protein, and fat) using milk analyzer (Milkoscope Julie C8 automatic, Scope Electric, Germany).

Statistical analysis

Data were statistically analyzed using the SAS System for Windows Release 9.0 (SAS Inst. Inc., 2002, Cary, NC). Body weights of ewes and lambs were analyzed according to the completely randomized design. The model contained the effects of dietary protein level, supplemental RPL and RPM, and the interactions between them. For lamb performance data, the initial BW of lambs was used as a covariate. Treatment means were computed using the LSMEANS option. Means were separated using pairwise comparisons of the least squares means using *t*-test.

Data for DMI, milk yield, and milk composition were analyzed as repeated measures according to the completely randomized design. The following model was used ($Y_{ijk} = \mu + T1_i + T2_j + T1_i * T2_j + W_k + T1_i * W_k + T2_j * W_k + T1_i * T2_j * W_k + E_{ijk}$, where Y_{ijk} = observation, μ = population mean, $T1_i$ = effects of dietary protein levels (i = 17 or 15), $T2_j$ = effects of supplemental L and M (j = yes or no), W_k = week effect (k = 2 to 5), and E = residual error. Milk yield for the first week was included as a covariate in the model for milk yield analysis. For each analysis, data was analyzed using the following covariance structures: compound symmetry, unstructured, autoregressive, and combination of structured and autoregressive. Subsequently, the appropriate covariance structure (smaller AIC and BIC values) for each analysis was selected. Treatment means were computed using the LSMEANS option and separated using preplanned pairwise comparisons of the least squares means using *t*-tests.

Results

Treatment diets

As planned, diets were isocaloric (averaged 9.75 MJ/kg of ME) but the MP diet contained lower CP (151 vs. 170 g/kg DM) and, subsequently, L (6.8 vs. 8.0 g/kg) than the HP diet (Table 1). Other nutrients were comparable in MP and HP diets.

Table 1 Ingredient and chemical composition of dietary treatments

Item	Dietary treatment[1]	
	HP	MP
Ingredient (g/kg DM)		
Barley	513	552
Wheat hay	244	254
Soybean meal	214	165
Limestone	17	17
Salt	11	11
Vitamin/mineral premix[2]	1	1
Nutrient (g/kg DM)		
Dry matter (DM)	902	903
Organic matter (OM)	901	902
Crude protein (CP)	170	151
Neutral detergent fiber (NDF)	339	350
Acid detergent fiber (ADF)	145	149
Ether extract (EE)	11	11
Lysine[3]	8.0	6.8
Methionine[3]	2.3	2.1
Metabolizable energy (ME; MJ/kg DM)[3]	9.8	9.7

[1] *HP*, high protein; *MP*, moderate protein

[2] Composition per 1 kg contained vitamin A, 2,000,000 IU; vitamin D3, 50,000 IU; vitamin E, 500 mg; vitamin C, 1000 mg; vitamin K_3, 20 mg; Ca, 200 gm, P, 80 gm; Mg, 40 gm; Fe, 500 mg; Zn, 2000 mg; Mg, 1000 mg; Cu, 300 mg; Se, 100 mg; I, 80 mg; and Co, 50 mg

[3] Based on tabular values (NRC, 2007)

Body weight of ewes and lambs

Initial and final BW of ewes and, subsequently, BW change of ewes were not affected by dietary treatments (Table 2). Initial BW of lambs was comparable among dietary treatments (Table 3). However, final BW, total gain, and ADG of lambs were higher ($P < 0.05$) when their dams were fed the MPYES diet compared to the MPNO and HPNO diets. For the HPYES diet, final BW, total gain, and ADG of lambs tended ($P \leq 0.10$) to be higher than those for the HPNO diet (Table 3).

Nutrient intake, milk yield, and milk composition of ewes

Effects of supplemental RPL and RPM on nutrient intake, milk yield, and milk composition of nursing ewes are presented in Table 4. There were no treatment × week effects ($P > 0.50$) for all data presented in Table 4. There were no interaction effects ($P \geq 0.88$) between dietary protein levels and supplemental RPL and RPM on nutrient intakes of ewes. Ewes fed the MP diets (MPNO and MPYES) had lower ($P < 0.01$) intake of CP (331 vs. 373 g/day) and L (15.7 vs. 18.2 g/day) but higher ($P < 0.01$) intakes of NDF (768 v.

Table 2 Effects of supplemental lysine and methionine on body weight of nursing ewes feed two levels of dietary protein

Item	Dietary treatment[1]				SEM	P-value[2]		
	HP		MP					
	No	Yes	No	Yes		CP * LM	CP	LM
n	9	9	8	8				
Initial body weight, kg	61.8	54.6	60.1	58.1	2.68	0.35	0.75	0.10
Final body weight, kg	60.9	55.5	58.0	56.5	2.43	0.44	0.71	0.17
Body weight change, kg[3]	−0.9	0.9	−2.1	−1.6	1.19	0.59	0.14	0.36

[1]Dietary treatments were arranged according to the 2×2 factorial design with two levels of protein (170 or 151 g/kg; HP or MP) and two levels of lysine plus methionine (0 or 8.5 plus 4.0 g/day/ewe, respectively; no or yes)

[2]*P*-values for effects of dietary protein (CP), supplemental lysine and methionine (LM), and interaction (CP * LM)

[3]Calculated as BW change = final BW − initial BW

Table 3 Performance of suckling lambs whose dams were fed diets with two levels of protein and supplemented with lysine and methionine

Item	Dietary treatment[1]				SEM	P-value[2]		
	HP		MP					
	No	Yes	No	Yes		CP * LM	CP	LM
n	9	9	8	8				
Initial body weight, kg	9.4	8.4	8.4	8.3	1.16	0.70	0.64	0.67
Final body weight, kg	15.0[b]	15.8[a,b]	15.3[b]	16.5[a]	0.35	0.58	0.17	<0.01
Total gain, kg	6.4[b]	7.2[a,b]	6.7[b]	7.9[a]	0.35	0.58	0.17	<0.01
Average daily gain, g/day	189[b]	212[a,b]	196[b]	233[a]	10.2	0.58	0.17	<0.01

[a,b]Within row, means with different superscripts are different at P value < 0.05

[1]Dietary treatments were arranged according to the 2×2 factorial design with two levels of protein (170 or 151 g/kg; HP or MP) and two levels of lysine plus methionine (0 or 8.5 plus 4.0 g/day/ewe, respectively; no or yes)

[2]*P*-values for effects of dietary protein (CP), supplemental lysine and methionine (LM), and interaction (CP * LM)

744 g) and ADF (327 vs. 318 g) compared to those fed the HP diets (HPNO and HPYES) with similar intakes of DM, OM, EE, M, and ME. Supplemental RPL and RPM had no effects ($P \geq 0.55$) on nutrient intake of ewes except for higher intake of L (17.9 vs. 16.1 g/day) and M (5.8 vs. 4.8 g/day).

Interactions between dietary protein levels and supplemental RPL and RPM were statistically significant ($P \leq 0.03$) for milk yield, milk efficiency, and milk composition yield. Milk yield for ewes fed the MPYES diet was higher ($P < 0.05$) than those fed the MPNO and HPYES diets and tended ($P = 0.08$) to be higher that the HPNO diet. Milk yield for ewes fed the MPNO diet was not significantly different from those fed the HPYES or HPNO diets. Milk yield was lowest for ewes fed the HPYES diet. Milk efficiency was highest ($P < 0.05$) for ewes fed the MPYES diet, but similar ($P \geq 0.20$) among other diets with a tendency ($P = 0.10$) for the HPNO to be higher than the HPYES diets.

Dietary treatments did not have substantial effects ($P \geq 0.13$) on milk composition. However, milk composition yields were affected by dietary treatments in a pattern similar to the effects on milk yield. Milk SNF, fat, protein, and lactose yields of ewes fed the MPYES diet were significantly ($P < 0.05$) higher than those fed the HPYES diets and tended ($P \leq 0.10$) to be higher than those fed the MPNO and HPNO diets. Milk composition yield was highest for ewes fed the MPYES diet and lowest for the HPYES diet.

Discussion

Studies on the effects of supplemental RPL plus RPM on performance of small ruminants are limited and none on nursing Awassi ewes. Thus, direct comparison of our results with published reports is not feasible. Regarding the effects of supplemental RPL and RPM on production performance, inconsistent results have been repetitively reported in dairy sheep and cows (Ali et al., 2009; Kim and Lee, 2021).

Table 4 Effects of supplemental lysine and methionine on nutrient intake, milk yield, and milk composition of nursing ewes feed two levels of dietary protein

Item	Dietary treatment[1]				SEM	*P*-value[2]		
	HP		MP					
	No	Yes	No	Yes		CP * LM	CP	LM
n	9	9	8	8				
Nutrient intake, g/day								
DM	2186	2202	2182	2208	19.5	0.82	0.95	0.32
OM	1970	1984	1969	1992	17.7	0.82	0.85	0.32
CP	372^{a}	374^{a}	329^{b}	333^{b}	3.0	0.87	< 0.01	0.31
NDF	741^{b}	$747^{b,c}$	$764^{a,c}$	773^{a}	6.8	0.82	< 0.01	0.32
ADF	317^{b}	$319^{b,c}$	$325^{a,c}$	329^{a}	3.0	0.82	< 0.01	0.31
EE	24	24	24	24	0.2	0.95	0.71	0.47
Lysine	17.3^{b}	19.1^{a}	14.8^{d}	16.6^{c}	0.11	0.88	< 0.01	< 0.01
Methionine	4.8^{b}	5.6^{a}	4.7^{b}	5.8^{a}	0.05	0.90	0.14	< 0.01
ME, MJ/day	21.3	21.4	21.2	21.5	0.20	0.82	0.95	0.31
Milk, g/day	$1646^{a,b}$	1368^{c}	$1522^{b,c}$	1897^{a}	95.1	< 0.01	0.04	0.62
Milk efficiency (kg milk/kg DMI * 100%)	72.5^{b}	62.9^{b}	70.7^{b}	87.5^{a}	5.2	0.02	0.04	0.49
Milk composition, %								
Solid nonfat	11.7	11.5	11.8	11.7	0.10	0.59	0.13	0.21
Fat	10.4	10.0	10.0	10.6	0.35	0.16	0.81	0.84
Protein	4.3	4.3	4.3	4.3	0.04	0.74	0.56	0.76
Lactose	6.3	6.3	6.4	6.4	0.05	0.84	0.35	0.35
Composition yield, g/day[3]								
Solid nonfat	$184^{a,b}$	158^{b}	$183^{a,b}$	221^{a}	14.5	0.03	0.04	0.66
Fat	$166^{a,b}$	137^{b}	156^{b}	201^{a}	14.5	0.02	0.08	0.62
Protein	$68^{a,b}$	59^{b}	$67^{a,b}$	81^{a}	5.2	0.03	0.06	0.66
Lactose	$100^{a,b}$	87^{b}	$99^{a,b}$	121^{a}	7.5	0.03	0.04	0.54

[a–d]Within row, means with different superscripts are different at *P* value < 0.05

[1]Dietary treatments were arranged according to the 2 × 2 factorial design with two levels of protein (170 or 151 g/kg; HP or MP) and two levels of lysine plus methionine (0 or 8.5 plus 4.0 g/day/ewe, respectively; no or yes)

[2]*P*-values for effects of dietary protein (CP), supplemental lysine and methionine (LM), and interaction (CP * LM)

[3]Calculated as composition yield = composition percentage * milk yield

Treatment diets

Protein contents of the diets used in the current study are comparable to diets (with 140 to 180 g/kg CP) that are commonly offered to nursing/lactating Awassi ewes in Jordan and other countries (Awawdeh et al., 2009, 2015).

Body weight of ewes and lambs

Consistent with our results, decreasing dietary protein levels and/or supplemental RPL and RPM had no effects on BW change of nursing (Lynch et al., 1991) or lactating (Sevi, et. Al., 1998) ewes. Decreasing protein contents of our ewes' diet did not affect the growth performance of their lambs and this is consistent with previous report (Lynch et al., 1991) where dietary protein was reduced from 163 to 102 g/kg. In the current study, growth performance (final BW, gain, and ADG) of lambs was improved when their dams were supplemented with RPL and RPM particularly when they were fed the MP diet. This improvement in lamb performance could be due to more milk produced by their dams as demonstrated in the current study and/or higher contents of L and M in their dams' milk (Lynch et al., 1991) or ration. Similarly, Lynch et al. (1991) observed improvement in BW gain and N balance of lambs that were nursing ewes fed diets supplemented with RPL and RPM regardless of protein levels (102 or 163 g/kg) of their dams' diets.

Nutrient intake, milk yield, and milk composition of ewes

Similar to our results, decreasing dietary protein levels (from 163 to 102 g/kg) had no effects on DMI of ewes (Lynch et al., 1991) but decreased CP intake, simply due to lower protein contents of the diets.

The current study demonstrates that decreasing dietary protein from 170 to 150 g/kg did not negatively impact milk yield and composition of nursing ewes. Consistent with our results, decreasing dietary protein from 163 to 102 g/kg had no effect on milk yield of black-faced ewes nursing twins (Lynch et al., 1991). Similarly, milk yield of lactating Comisana ewes was not reduced with decreasing dietary protein from 157 to 129 g/kg (Sevi et al., 1998). It has been demonstrated that the effect of dietary protein level on productivity of dairy cows is inconsistent (Huhtanen and Hristov, 2009). The fact that milk yield of our ewes did not increase with increasing dietary protein indicates enough protein (AA) supply from the diet and/or greater contribution of body protein reserve to milk synthesis in ewes fed the MP diet. It has been suggested that the portion of N used for milk synthesis increases in ewes fed low-protein diets (Lynch et al., 1991; Sevi et al., 1998).

Based on recorded average DMI and milk yield, our ewes that were fed the HP diet had about 123% of the recommended metabolizable protein intake (NRC, 2007) and those fed the MP diet had about 100%. There has been growing interest to reduce protein contents of ruminant diets without negatively affecting the quality of metabolizable protein (by using ruminally protected AA) and, subsequently, animal performance (Sinclair et al., 2014; Abbasi et al., 2019). Our results suggest that dietary protein for ewes similar to those used in the current study can be reduced to, or probably below, 150 g/kg without affecting their performance.

In the current study, supplemental RPL and RPM improved milk yield of ewes fed the MP diet. When ewes were fed the HP diet, milk yield was not improved in response to RPL and RPM supplementation and actually it was reduced. Although we do not have a definite explanation which deserves further investigation, this reduction is perhaps due to imbalanced AA in absorbable protein that could have led to inefficient utilization of AA for milk protein synthesis (Sevi et. al., 1998). Robinson et al. (2000) reported that production (milk, protein, and lactose yields) was depressed when dairy cows were abomasally infused with excess (140% of calculated requirement) lysine and methionine. Our results demonstrate that balancing rations with RPL and RPM improved protein utilization for milk synthesis when ewes were fed diets supplying adequate (MP diet), but not excess (HP), metabolizable protein. On the other hand, Lynch et al. (1991) reported that RPL and RPM failed to improve milk yield of nursing ewes fed diets deficient in protein supply.

In contrast to our results, supplemental RPL and RPM improved milk yield of Comisana ewes (Sevi et al., 1998) regardless of dietary protein levels (157 or 129 g/kg) and failed to improve milk yield of black-faced ewes (Lynch et al., 1991) at both dietary protein contents (163 or 102 g/kg). Recently, RPL or/and RPM supplementation did not improve milk yield of Chios ewes fed diets containing 139 g/kg CP (Mavrommatis et al., 2021). These discrepancies could be attributed to many factors such as composition of the basal diets, type and level of supplemental AA, level of production, and ewe breed.

The highest milk yield in our study was for ewes fed the MP diet and supplemented with RPL and RPM. This suggests a better utilization efficiency of dietary protein. This is in agreement with the findings in Comisana ewes where the highest gross utilization efficiency of protein was observed in ewes supplemented with RPL and RPM and receiving the low (129 g/kg) compared to high (157 g/kg) protein diets (Sevi et al., 1998). This supports the important concept of balancing AA (mainly L and M) in metabolizable protein where protein utilization by the mammary gland can be improved as a result of a better partitioning of nutrients to the mammary gland.

As for milk yield, the effects of dietary protein levels and supplemental RPL and RPM on milk composition have been variable. In the current study, dietary protein levels and/or supplemental RPL and RPM did not have substantial effects on milk composition. In contrast, decreasing dietary protein levels reduced milk protein contents of ewes (Lynch et al., 1991; Sevi et al., 1998) with no effects on milk fat contents (Sevi et al., 1998). Relatively, our moderate-protein diet (151 g/kg) contained higher protein than the low-protein diets (102 and 129 g/kg, respectively) reported in other studies (Lynch et al., 1991; Sevi et al., 1998). This could be the reason for not observing depression in milk protein contents in response to deceased dietary protein levels as in the previous studies (Lynch et al., 1991; Sevi et al., 1998).

It has been reported that supplemental RPL and RPM improved milk protein and fat contents of Comisana ewes fed either low (129 g/kg) or moderate (157 g/kg) protein diets (Sevi et al., 1998), and increased milk protein contents of black-faced ewes fed low (102 g/kg) but not moderate (163 g/kg) protein diets (Lynch et al., 1991). Recently, milk fat but not protein contents were improved with RPL and RPM supplementation in dairy Chios ewes fed moderate (139 g/kg) protein diets (Mavrommatis et al., 2021). It is obvious that the effects of supplemental RPL and RPM on milk composition are inconsistent and may be affected by several factors similar to those that affect the response in milk yield.

Conclusions

Under conditions similar to our study, dietary protein can be reduced from 170 to 151 g/kg without negatively impacting the performance of ewes (nutrient intake, milk yield, and milk composition) and growth performance of their lambs. The effect of supplemental RPL and RPM on milk yield of ewes is dependent on protein contents of the offered diet. Milk yield, milk composition yield, and milk efficiency of ewes and growth performance of their lambs were improved with supplemental RPL and RPM when ewes were offered relatively moderate-protein, but not high-protein, diets. In fact, if nursing ewes are offered high-protein diets, supplemental RPL and RPM may be disadvantageous. Further studies are needed to determine the optimal dietary protein contents and amounts of supplemental RPL and RPM at different production stages of ewes.

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Author contribution All roles (conceptualization, funding acquisition, methodology, data curation, formal analysis and investigation, supervision, and manuscript writing) were performed by the corresponding author (M. S. Awawdeh).

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Data availability Data generated during this study are included in the current article.

Declarations

Ethics approval Not applicable.

Consent to participate Not applicable.

Consent for publication Not applicable.

Code availability Not applicable.

Conflict of interest The author declares no competing interests.

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