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Research Article

Postpartum Associated Metabolism, Milk Production and Reproductive Efficiency of Barki and Rahmani Subtropical Fat-tailed Breeds

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Abstract

This study aimed to evaluate postpartum associated metabolism as an adaptive indicator for lactational stress, milk production and reproductive efficiency of two subtropical sheep breeds. Blood samples were collected from Rahmani (n = 15) and Barki (n = 15) ewes, 14 days postpartum and at biweekly interval thereafter (until day 56 postpartum). Milk yield and milk composition were determined and were used to calculate energy corrected milk, milk energy value and energy required for lactation. Ovarian activity was biweekly monitored using transrectal ultrasonography until day 56 postpartum and then ewes were synchronized for estrus 60 days postpartum and were mated. Rahmani ewes had greater ($p<0.05$) overall concentrations of serum glucose, total protein and insulin than Barki ewes. No differences were observed between the two breeds in the concentrations of serum urea and T_3 hormone. Milk yield of Barki ewes was higher ($p<0.05$) than Rahmani ewes, but energy corrected milk was in the same trend. Further, Rahmani ewes tended ($p = 0.09$) to produce milk with higher energy value. The calculated net energy required for lactation was in the same average in both breeds. No differences in milk composition were observed in both breeds, although, percentage of total solids was higher ($p = 0.07$) in Rahmani ewes. Following estrous synchronization, Rahmani ewes had higher ($p<0.05$) number of corpora lutea 11 days post-mating and lower embryonic loss ($p = 0.14$), also they had higher conception ($p = 0.09$) and fecundity ($p<0.02$) rates compared with Barki ewes. In conclusion, postpartum metabolic profile could be used as an indicator on breed superiority to subsequent productive and reproductive performance.

Key words: Blood metabolites, breed, postpartum, fertility, subtropical sheep

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INTRODUCTION

Rahmani and Barki fat-tailed sheep are considered to be the most predominant breeds in many Middle East countries (Galal *et al*., 2005). These breeds are mainly kept for meat production (Abdel-Moneim, 2009; Hashem *et al*., 2015). Thus, lamb crop is an important economic trait which could be maximized by applying the accelerating-lamb production system, three successive lambings in 2 years. In this system, female sheep are bred within 60 days postpartum during which the peak of lactation occurs (Aboul-Naga *et al*., 1981). Lactation is a physiological event associated with many changes in metabolic profile to ensure the nutrients supply for milk production (Krajnicakova *et al*., 2003; Kashinakunti *et al*., 2010). Normal blood levels of various metabolites are important for normal physiological functions including reproductive system (Blache *et al*., 2008). Blood glucose, cholesterol and protein are some of nutrients affecting fertility and cyclicity in farm animals (Qureshi, 1998; Park *et al*., 2007; Karapehlivan *et al*., 2007). Such blood metabolites signal the amount of energy that is readily available from digestion and from body stores, in addition to their direct influences on the hypothalamo-pituitary-ovarian axis (Martin *et al*., 2004). Previous studies, reported that different breeds of sheep and goats exert varied degrees of metabolic adaptation to lactational stress (Abdelrahman and Aljumaah, 2012; Anwar *et al*., 2012) which is genetically controlled (Roubies *et al*., 2006). Measuring blood biochemical attributes during lactation is an important indication of the degree of tissue damage and evolutionary adaptation of the animal to lactation (Gupta *et al*., 2007; Anwar *et al*., 2012). Therefore, the objective of the present study was to evaluate postpartum associated metabolism, milk production and reproductive potential of Barki and Rahmani subtropical fat-tailed ewes in early to mid-lactation.

MATERIALS AND METHODS

Animal's care and management: The present study was conducted at the Agricultural Experimental Station (31°20′N, 30°E), Faculty of Agriculture, Alexandria University. All the procedures applied on the animals were carried out in accordance with the Guide for the Care and Use of Agricultural Animals in Research and Teaching, Federation of Animal Science Societies (FASS). Thirty multiparous, lactating Rahmani and Barki ewes at 3-6 years of age, weighing 47.50±1.78 and 44.74±1.78 kg at allocation (day 14) were used in the present study. All ewes were kept outdoors with shelter during the day and housed in a semi-open barn at night. All ewes were fed a commercial concentrate mixture (63% TDN and 14% CP) and a green clover (*Trifolium alexandrinum*) according to NRC (2001). Animals had free access to fresh tap water during the whole experimental period. The ewes were disease-free with a healthy appearance. Ewes were weighed twice on days 14 and 60 postpartum.

Blood sampling and analyses: Samples of 10 mL of blood were collected by means of jugular venipuncture in non-heparinized tubes. The blood samples were collected from each ewe on days 14, 28, 42 and 56 postpartum. All samples were collected before the morning fed ration. Serum was separated by centrifugation of samples at 700×g for 20 min and was frozen at -20°C for later analyses. Concentrations of serum glucose, triglycerides, total protein and urea were determined using commercial kits (Stanbio, Texas, USA.). Additionally, concentrations of serum insulin and triiodothyronine (T_3) were also measured using commercially available soiled-phase enzyme immunoassay kits (DRG International, USA). Sensitivities of the methods were 1.76 μlU mL^{-1} and 0.2 ng mL^{-1} for insulin and T_3, respectively. The corresponding intra and inter-assay coefficients of variations were 2.1–5.7 and 5.6-5.0%, respectively.

Determination of milk yield and milk composition: Milk yield was determined biweekly starting 14 days postpartum. Lambs were removed from their dams and kept in separate pens 20 h prior to milking process. Milk yield was determined by milking out the ewes by hand and weighed thereafter. Values obtained were multiplied by a factor of 1.2 to get the milk yield for 24 h. Samples of 50 mL were immediately collected and analyzed for milk composition by Milk Analyzer (Master ECO., Bulgaria). The yield of Energy Corrected Milk (ECM) was calculated according to the equation: ECM (g day^{-1}) = milk yield (g day^{-1})×[(0.0929×% of milk fat)+(0.0563×% of milk crude protein)+(0.0395×% of milk lactose)] /0.749 Mcal kg^{-1} (NRC., 2001). Milk energy value (MEV, kcal kg^{-1}) was calculated according to Baldi *et al*. (1991) as 203.8+(8.36×% of milk fat)+(6.29×% of milk crude protein). Net energy required for lactation (NEL; Mcal kg^{-1}) was calculated as milk yield (kg)×[(0.0929×% of milk fat)+(0.0547×% of milk crude protein)+(0.0395×% of milk lactose)]. The NE required for maintenance (NEM; Mcal day^{-1}) was calculated as $BW^{0.75x}$ 0.08 (NRC., 2001).

Assessment of ovarian activity and estrous synchronization: The ovaries of each ewe was biweekly monitored on days 14,

28, 42 and 56 postpartum using an ultrasound scanner equipped with 5 and 7.5 MHz linear array B-mode real-time endorectal probe (Pie Medical Equipment B.V., Maastricht, Netherlands). Animals were banned from feed for 12 h before examination, which was carried out in a standing position. The probe was fitted to a plastic rod (1×30 cm) as an adapter to enable the insertion of the probe into the rectum. The probe was lubricated by a hydro soluble gel and sheathed with polyvinyl chloride pipe (2×35 cm) to avoid damage of the rectal mucosa and was gently inserted about 20 cm through the rectum after feces removal until the anechoic content of the bladder was visible on the screen and then the probe was rotated 90° clockwise and 180° counterclockwise across the reproductive tract until the uterine horns and both ovaries were scanned. Presence of the corpus luteum in the ovaries was used to estimate percentage of ewes had ovulated and also time required for resumption of ovarian activity. On day 60 postpartum, ewes were subjected to estrous synchronization by feeding ewes 0.3 mg of melanogesterol acetate (MGA, Pharmacia Animal Health, Kalamazoo, MI) for a period of 7 days and administered 0.5 mL of prostaglandein $F_{2\alpha}$ ($PGF_{2\alpha}$, Estrumate, 250 μg cloprostenol/mL, Schering-Plough Animal Health, Germany) at the last day of MGA feeding. This was followed by 1 mL of gonadotropin releasing hormone (GnRH, Receptal®, 0.004 mg buserelin, Boxmeer, Holland) 4 days later and a second $PGF_{2\alpha}$ injection 7 days after GnRH injection. The protocol is known as MGA: 7-11Synch by Kojima *et al*. (2000) to synchronize estrus and ovulation. One day following the end of estrous synchronization protocol, the overt signs of estrus were detected twice daily for 1h using teaser rams for 1 week. Ewes displaying signs of estrus were subjected to natural assisted mating by proven fertile rams every 12 h until the disappearance of estrous signs. Also, numbers of Corpora Lutea (CLs) were recorded on days 11 post-mating. The CLs detected on day 11 post-mating were expressed as ovulation. Pregnancy diagnosis and number of embryos per each ewe were recorded by scanning the uterine contents at 35 days post-mating.

Statistical analysis: Statistics of the serum metabolism, milk yield and milk composition were carried out using a linear mixed effects model (PROC MIXED from SAS version 9.1) (SAS., 2004) for repeated measurements, including breed (Rahmani and Barki) and sampling time (14, 28, 42 and 56) as fixed factors and the ewes effect as a random factor. The same procedure was used for the animal body weight but on two measuring times (day 14 and 60 postpartum). Categorical data was analysed using chi-square test. Differences between the means in both breeds were tested by Duncan's multiple range test. All results were expressed as Least Square Mean (LSM)±Standard Error of Mean (SEM). The statistical significance was accepted at $p \leq 0.05$.

RESULTS

Body weight and blood metabolites of Barki and Rahmani ewes: Data for body weight and metabolic profile of Rahmani and Barki ewes throughout 60 days postpartum are presented in Table 1 and 2. Rahmani ewes had ($p<0.001$) higher overall mean of body weight than that of Barki ewes. From day 14 to 60 postpartum, body weights decreased 2.47 and 2.75 kg in Rahmani and Barki ewes, respectively; however, this loss did not result in a significant change in the body weight of ewes during the experimental period (day 60 postpartum; Table 1). Net energy required for maintenance (Mcal day^{-1}) was significantly higher in Rahmani ewes than in Barki ewes (Table 1). Rahmani ewes had greater ($p<0.05$) overall concentrations of serum glucose, total protein and insulin than Barki ewes. Also, Rahmani ewes tended ($p=0.106$) to had higher concentration of triglycerides than Barki ewes (Table 2). No differences were observed between the two breeds in the concentrations of serum urea and T_3 hormone (Table 2).

Milk yield and composition: Milk yield and its composition were significantly affected by breed (Table 3). Milk yield of

Table 1: Least square mean (±SEM) of body weight of postpartum Rahmani and Barki ewes

	Breed			
Parameters	Rahmani	Barki	SEM	p-value
Body weight (BW) (kg)	46.26[a]	43.37[b]	1.47	0.007
BW on day 14 postpartum	47.50	44.74	1.78	0.889
BW on day 60 postpartum	45.00	42.00	1.78	0.889
NEM (Mcal day^{-1})	1.39[a]	1.32[b]	0.022	0.054
Weight change (kg)	2.47	2.75	0.63	0.648

[a,b]Means with different superscripts within the same row differ significantly ($p<0.05$) and NEM: Net energy required for maintenance

Table 2: Least square mean (±SEM) of blood serum biochemicals in postpartum Rahmani and Barki ewes throughout the experimental period

	Breed			
Parameters	Rahmani	Barki	SEM	p-value
Insulin (μlU mL^{-1})	32.33[a]	25.01[b]	2.50	0.034
T_3 (ng mL^{-1})	0.538	0.607	0.03	0.196
Glucose (mg dL^{-1})	75.30[a]	71.71[b]	1.91	0.002
Triglycerides (mg dL^{-1})	8.16	7.35	0.83	0.106
Total protein (mg dL^{-1})	7.42[a]	7.04[b]	0.12	0.020
Urea (mg dL^{-1})	74.04	72.88	1.48	0.583

[a,b]Means with different superscripts within the same row differ significantly ($p<0.05$)

Table 3: Milk yield and composition of Rahmani and Barki fat-tailed sheep (LSM±SEM)

	Breed			
Parameters	Rahmani	Barki	SEM	p-value
Milk yield (g day^{-1})	455.64^{b}	508.92^{a}	15.0	0.046
Energy corrected milk yield (g day^{-1})	493.80	534.00	17.1	0.209
Milk energy value (kcal kg^{-1})	262.06	259.10	1.41	0.09
Energy for lactation (Mcal kg^{-1})	0.819	0.786	0.228	0.230
Milk composition (%)				
Fat	3.47	3.15	0.14	0.132
Protein	4.64	4.59	0.03	0.394
Lactose	5.93	5.93	0.05	0.935
Total solids-not-fat	13.60	13.53	0.08	0.675
Total solids	17.83	17.32	0.16	0.078

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

Table 4: Reproductive performance of Barki and Rahmani ewes bred within 60 days postpartum

	Breed			
Parameters	Rahmani	Barki	SEM	p-value
No. of ewes	15	15	-	-
Percentage of ewes had ovulated[1]	66.6 (10/15)	73.3 (11/15)	-	0.847
Days for first detected CL[1]	48.00	48.00		11.47
Estrus rate[2] (%)	80.0 (12/15)	73.3 (11/15)	-	0.715
No. of CL[3]/ewe	1.13^{a}	0.93^{b}	0.08	0.046
Diameter of CL[3] (mm)	10.74^{b}	12.03^{a}	0.310	0.019
Embryonic loss[2] (%)	25.00	54.55	-	0.141
Conception rate[2] (%)	75.0 (9/12)	45.4 (5/11)	-	0.091
Fecundity rate[2] (%)	91.6^{a} (11/12)	45.4^{b} (5/11)	-	0.028

[1]Parameters were recorded before estrous synchronization (days 14, 28, 42 and 56 post-partum), [2]Estrus rate = (No. of ewes displayed estrus/No. of induced ewes)×100%, conception rate = (No. of ewes conceived on day 35/No. of ewes exposed)×100%, embryonic loss = (No. of detected embryos on day 35/No. of counted CL on day 11 post-mating) and fecundity rate = (No. of lambs born/No. of ewes exposed)×100% and [3]Data of numbers and diameters of corpora lutea were taken on day 11 of the induced luteal phase

Barki ewes was higher than Rahmani ewes, however this difference was not observed when the milk yield was corrected for energy. Further, Rahmani ewes tended ($p = 0.09$) to produce milk with higher energy value. The calculated net energy required for lactation (Mcal kg^{-1}) was in the same average in both breeds. Percentages of milk fat, protein, lactose and solids-not-fat were not different in both breeds. Percentage of total solids was higher ($p = 0.07$) in Rahmani ewes compared to Barki ewes.

Reproductive performance following estrous synchronization: Resumption of ovarian activity and reproductive performance of Rahmani and Barki ewes following estrous synchronization is set out in Table 4. Resumption of ovarian activity (first detected CL postpartum) occurred 48.0±11.47 days postpartum in 66.6% of Rahmani ewes and 73.3% of Barki ewes (Table 4). Rahmani ewes had higher ($p<0.05$) No. of corpora lutea 11 days post-mating with lesser ($p<0.05$) diameters than Barki ewes. Also, Rahmani ewes tended to have lower embryonic loss ($p = 0.14$), also they had higher conception ($p = 0.09$) and fecundity ($p<0.02$) rates compared with Barki ewes.

DISCUSSION

In this study, both sheep breeds did not loss significantly their body weights. This indicates that the animals received adequate nutrients and the variations observed between the two breeds in the concentrations of the blood metabolites were attributed mainly to the breed effect. In this study, Rahmani ewes had better metabolic status than Barki ewes during 60 days postpartum, indicating higher metabolic adaptation of this breed to lactational stress. This result is consistent with that of Abdelrahman and Aljumaah (2012) who found variations in blood metabolites of Najdi and Naemi sheep at parturition and early lactation. In context, Anwar *et al*. (2012) found that among four goat breeds (Anglo-Nubian, Angora, Baladi and Damascus), Baladi goats (an Egyptian breed) had the most adaptive characteristics during the first week of lactation, where they showed the highest concentrations of serum total protein, cholesterol, total lipids and glucose compared with other breeds.

During lactation the mammary gland is considered the major energy consuming organ, it utilizes most of the circulating metabolites in the blood for milk synthesis (Karapehlivan *et al*., 2007). Therefore, lactating females develop different adaptation mechanisms to offer different nutritional precursors required for milk production (Emmison *et al*., 1991). One of these mechanisms is the increased efficiency of gluconeogenesis process in the hepatocytes, whereas it reaches its maximum rate in hepatocytes isolated from lactating ewes compared with non-lactating ewes. It was reported that in sheep, high concentrations of insulin increase the rate of gluconeogenesis from propionate in hepatocytes isolated from lactating animals which enables the animal to cope with the increased demand for glucose by the mammary gland (Emmison *et al*., 1991). In the present study, Rahmani ewes had higher level of serum insulin which according to previous facts could be the reason for better hepatic gluconeogenesis and thus improved glucose level in this breed. Additionally, higher levels of serum glucose could be due to the increased insulin resistant in muscles and adipose-tissues which reflects other adaptation mechanism in this breed during lactation. It could be also

observed that Rahmani ewes had higher levels of serum total protein and triglycerides during their postpartum period. Both breeds used in this study are classified as fat-tailed breeds, however Rahmani sheep have been known to have heavier fat-tail (Abdel-Moneim, 2009). The fat-tail is regarded as an adaptive response of animals to a hazardous environment and is a valuable reserve for the animal to be utilized during pregnancy and lactation (Kashan *et al*., 2005). These facts present another adaptive characteristic of this breed to lactational stresses and their ability to compensate different blood metabolites efficiently during postpartum period than Barki ewes.

In the present study, better serum metabolites were associated with adequate subsequent milk and reproductive performance. Comparing the milk production and reproductive performance of both breeds investigated in this study. It is clear that, however, Barki ewes produced higher milk yield, this different disappeared when milk yield was corrected to energy and also the energy value of the milk produced by Rahmani ewes was higher than that of Barki ewes. Considering that both energy corrected milk yield (that considers the percentages of milk fat, protein and lactose) and milk energy value reflect the energy output from animal body (Linn, 2003). On the other hand, net energy for lactation indicates the feed energy available for milk production after digestive and metabolic losses required for milk production, was not different between the two breeds in this study. Accordingly, it could be said that Rahmani ewes were more efficient in energy utilization than Barki ewes.

In this study, Rahmani and Barki ewes resumed their ovulation activity within 48 days postpartum. This value is in accordance with that previously reported in sheep (Hayder and Ali, 2008). It could be observed that, however, both breeds resumed their ovarian activity within the same time, only Rahmani ewes showed adequate reproductive performance (higher conception and fecundity rates). Rahmani ewes was reported to have higher ovulation rate and litter size than other endogenous sheep breeds including Barki ewes (Abdel-Mageed and Abo El-Maaty, 2012; Hashem and El-Zarkouny, 2014). These findings are in accordance with the present study where Rahmani ewes produced more lamb crop than Barki ewes. This could be mainly ascribed to the fact that Rahmani ewes had higher number of corpora lutea (ovulation rate) and lower embryonic loss than Barki ewes. Higher embryonic viability in Rahmani sheep may reflect the ability of this breed to partition nutrients toward the uterus, supporting embryonic and/or fetal growth.

CONCLUSION

Rahmani ewes exerted better postpartum adaptive characteristics as denoted by higher productive and reproductive capacity which makes it a suitable breed for intensive production system. Postpartum metabolic profile could be used as an indicator on breed superiority to subsequent productive and reproductive performance. More studies are required to understanding the adaptability aspects of different subtropical endogenous sheep breeds for achieving maximum productivity during their production life time.

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