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Effects of calcium salts of fatty acids (Megalac) on reproductive performance and blood parameters of Kalkohi ewes

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Abstract

To determine the effects of Megalac addition on Iranian ewes' reproductive parameters, a complete randomized design was used. Multiparous Kalkohi ewes (n = 32; 3 years old) were randomly assigned to 2 groups which consumed isoenergetic and isonitrogenous control (C) or Megalac (M; 5% Megalac in diet DM) diet. All ewes were synchronized by prostaglandin injection. Blood samples were collected from d 8, 10, and 12 of estrus cycle. Diets offered 4 wk prior to mating and up to 4 wk after mating and 2 weeks before parturition. Pregnancy, lambing and twining rate were not significantly affected by fat inclusion in ewe diet ($P \geq$ %5). Similarly, pregnancy length was unaltered by treatment ($P \geq$ %5). Lambing crop was affected by diet fat ($P \leq$ %5). Cholesterol concentration significantly increased as Megalac added to the diet (52.7 vs. 49.7 mg/dl in M and C, respectively; $P \leq$ %5), whereas P4 concentration was similar between M and C group in d 8, 10 and 12 of estrus cycle (3.80 and 3.81 nmol/L for M and C, respectively; $P \geq$ %5). This experiment suggests that fatty acid content and profile of this level of Megalac may be insufficient for improving ewes' reproduction performance, but 2.5 % Megalac in ewes' diet DM before parturition could improve lamb weight. The lack of changes in P4 concentrations alongside the increasing of blood cholesterol did not support cholesterol and P4 relation. However, further studies are needed to determine impact of various fat sources and levels on physiological responses in ewes.

Key words: Megalac, reproduction, cholesterol, progesterone, ewe

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Introduction

Many researchers are trying so hard and believe by improvement of nutrition conditions it is possible to improve ewe's reproduction and adding Megalac is an effective way to reach this goal (Hess et al., 2008). However, due to limited information regarding determining desired level and appropriate type of fat it requires lots of studies in sheep's nutrition. The obtained answer after using Megalac in ruminant diet were different and in some cases improvement of reproduction of performance due to adding Megalac with high conception rate or decreasing of open days were observed (Sklan et al., 1988), On the other hand, there are some experiments that show there is no significant changes in reproduction performance regarding to addition of fat supplements (Caroll et al., 1990).

There are different theories about this matter, among them maybe mentioned the following: synthesis increase in steroid compounds like steroid hormones amongst progesterone, concentration change of blood insulin followed by stimulation growth of ovarian follicles and decrease in PGF2 synthesis which this compound has negative effect on corpus luteum activity (Staples et al., 1998).

Rumen protected fat production is mainly in order to deal with negative effect of fat on fiber digestion, prohibiting biohydrogenation and increasing long chained fatty acids to small intestine (Hess et al., 2008). This method was provided some years ago and the major advantage of it is the neutral fatty calcium soap consumed in rumen and improves the level of essential fatty acids. The researchers showed that 40 grams of protected fat on Ghezel breed ewe's diet per day will increase the number of ovarian follicles larger than 7 mm in diameter (Zamiri et al., 2002). In the same way researchers reported with the fish oil feeding the total number of follicles in ewes fed with diet which contains 4% fish oil calcium salt was more comparing to the other group (Sklan et al., 2002). It was shown in Zandi ewes that conception rate will improve in control group if we consume 4.5% calcium salt of soya fatty acid instead of 4.5% calcium salt of tallow mixture fatty acid and also calcium salt from mixture of tallow and soya in their diet (Sadeghipanah, 2002).

Observing the level of hormones is very important in reproduction. The level of blood progesterone is one of the most important factors in maintaining pregnancy.

According to researchers a physiological mechanism is responsible for increasing of progesterone concentration in serum due to decreasing of removal rate of progesterone from blood or likely the concentration increase of its precursor (cholesterol) (Hawkings et al., 1995). In a research with daily nutrition of 50 grams of fatty acid calcium salt there was no change between progesterone concentration in ewes blood in days 6 and 12 of the first reproduction cycle postpartum (Espinoza et al., 1998). But in total there are conflicting results from changes and correlations of cholesterol with blood progesterone is observed. Conclusion of nutritional observations shows that it is possible for fat resources to improve reproduction, but in the case of Iranian sheep few studies have been done. Also,

effects of consuming fat supplements on blood parameters and relevancy of cholesterol and progesterone is still controversial.

Therefore, present experiment is to determine the effects of fatty acid calcium salts (Megalac) on reproductive performance and blood parameters (progesterone and cholesterol) of Kalkohi ewes in the breeding season was performed.

Materials and Methods

This experiment has been done in research and educational farm of Islamic Azad University of Saveh by using 32, three years old non-pregnant, non-lactating Kalkouhi ewes with the average weight of 52.3 Kg in completely randomized design from late August 2008 till January 2009.

In order to investigate the effects of fatty acid calcium salts (Megalac) on reproductive characteristics and blood parameters of Kalkouhi ewes they were separated to two Control and Treatment (Megalac Consumer) groups in boxes of four ewes each. On Treatment group 5% of DM was considered to be Megalac, all the diets was based on NRC 1985 so in terms of energy and protein they were similar (Table 1).

All the feeds were given four week prior and after the experiment period twice a day (9 am and 3 pm). In order to making the ewes compatible to the feed containing Megalac it takes about a week to reach the amount that we want to, which is 5% in DM's diet. After 8 weeks of feeding the ewes and having the rams in, all the experimental livestock joined the main herd and they were treated same as the herd (in the morning: Wheat Straw, Barley, Wheat Bran and at noon: Alfalfa) for 12 weeks until the possible date of parturition. Two weeks before the start of possible parturition date both control and treatment (5.2% fat in DM) were separated from the herd and they were fed as Table 2. It should be noted that also in this period compatibility of Megalac was observed (Table 2).

The studied factors were as following: A) Pregnancy ratio: Percentage of pregnant ewes to the total number of ewes, B) Twining ratio: Percentage of biparous and multiparous ewes to the number of ewes, C) Lambing ratio: Percentage of born lambs to total number of ewes, D) Lamb production: weight of born lamb for each ewe to the length of pregnancy period. Regarding to heat synchronization two rounds of 1ml Prostaglandin (PGF2α) with the market name of Vetaglandin (D-Cloprostenol, Aburaihan Pharmaceutical Co., Tehran, Iran) were given within 9 days in Biceps femoris.

The first heat cycle was diagnosed with heat detector rams that is why 40X60piece of cloth was fastened under the stomach of the ram due to mating prevention. The first heat cycle was considered in order to collect the blood samples and determine levels of serum progesterone and cholesterol, also

Table 1. Ingredient and chemical composition of the diets during mating period (% of DM basis).

Item	Value	
	Control	Treatment
Ingredient		
Alfalfa	45.0	50.0
Wheat straw	28.0	27.8
Barley grain	26.0	16.0
Vitamin and mineral mix	1.0	1.0
Dicalcium phosphate	-	5.0
Fat supplement (Meglac)	-	0.2
		5.0
Chemical composition[1]		
ME (Mcal/kg)	2.16	2.16
TDN	0.60	0.60
CP	10.0	10.0
Ca	0.70	1.20
P	0.20	0.20

[1]TDN = total digestible nutrients; CP = crude protein

Table 2. Ingredient and chemical composition of the diets during two weeks before lambing (% of DM basis).

Item	Value	
	Control	Treatment
Ingredient		
Alfalfa	29.2	25.6
Wheat straw	31.5	36.0
Barley grain	32.7	25.0
Wheat bran	9.70	5.00
Vitamin and mineral mix	1.0	1.0
Dicalcium phosphate	0.20	0.20
Calcium carbonate	0.40	-
Fat supplement (Meglac)	-	2.50
Chemical composition[1]		
ME (Mcal/kg)	4.10	4.15
TDN	1.10	1.10
CP	9.60	9.50
Ca	0.60	0.70
P	0.30	0.30

[1]TDN = total digestible nutrients; CP = crude protein

considering next heat cycles for mating. Blood sampling was performed 3 times a day, 2 hours after feeding in the morning on 8, 10, 12 days after the first estrous cycle from the Jugular vein. Blood samples were centrifuged for 5 min in 4500 rpm and serums were frozen in -20`C. All data were statistically analyzed using GLM procedure of SAS. Observations averages were compared with 5% error level by Duncan's multiple range tests.

Results

The effects of fatty acid calcium salts consumption are shown in Table 3. Using fat supplement at ram releasing period has no effects on pregnancy ratio, lambing ratio, twining ratio or length of

Table 3. Effect of fat supplementation on reproductive performance in ewes.

Parameters	Experimental Groups		*P*-value
	Control	Treatment	
Pregnancy rate (%)	92.0	100.0	ns
Lambing arte (%)	138.0	167.0	ns
Twinning rate (%)	38.0	50.0	ns
Lambing crop (kg)	4.68	6.31	*
Gestation period (day)	151.0	150.4	ns

Table 4. Blood metabolites of experimental ewes during 8, 10, and 12 days of estrus cycle (mean ± SD).

Parameters	Experimental Groups		*P*-value
	Control	Treatment	
Cholesterol (mg/dl)	49.7 ± 2.25	52.7 ± 4.65	*
Progesterone (nmol/l)	3.81 ± 0.59	3.80 ± 0.66	ns

pregnancy period ($P > 0.05$). However fat consumption 2 weeks before birth has significantly improved the lamb production ($P < 0.05$).

Changes in blood parameters were significant, by consumption of 5% fatty acid calcium salt the level of ewes' blood cholesterol was significantly higher, comparing to control group (7.52 vs. 7.42 mgr/dl for treatment and control groups, respectively). On the other hand, there is no significant change in blood progesterone levels from days 8 to 12 of estrus cycle.

Discussion

There was no consistency about the positive effect of Megalac on pregnancy ratio with Sadeghipanah (2002) finding regarding consumption of soy bean fatty acid calcium salts. It seems the difference between the results is because of different source of consumed fatty acid. In terms of quality soy bean is much better than Megalac (omega 6 source) and maybe this is the reason why there was no positive effect.

Also in the above experiment there are significant differences between tallow fatty acid calcium salts and mixture of tallow and soy bean seen that shows significant effects of fat source.

There was no difference between two groups in twining and lambing ratios. These results are consistent to (Zamiri et al., 2002) observations. The mechanism of how fat effects number of follicles is not clear, however it is as fat may effect central nervous system and GnRH excretion by metabolites and hormones. Fat may also effects follicles through hormones and metabolites activity on ovary surface (De Fries et al., 1998). Although no significant changes on blood progesterone levels could be in line with the same results.

No changes in length of pregnancy period is consistent to other experiments on Ghezel and Mehraban ewes and all researchers agree on this point that consumption of fat sources has no significant effect on length of pregnancy period (Ghoreishi, 1998). Feeding fat supplement increase the birth weight of lambs in treatment group and this is despite isoenergetic dietary intake in both groups. In a study on Holstein cows they did the same thing but there was no difference in calf weight (Lammoglia et al., 1996). They suggested that fat intake was not sufficient to stimulate fetal growth. But it seems that levels of fatty acid calcium salt were good enough to improve the birth weight of the lambs. By consumption of fat supplement in treatment group cholesterol concentration was increased. In different researches that they used fat in the feed of ruminants such as ewes, fat supplement increased the concentration level of blood cholesterol (Espinoza etal., 1998; Kuran et.al, 1999).

In vitro experiments show that synthesis of steroids in luteal tissues depends on the availability of cholesterol. So increasing of cholesterol can increase progesterone production in follicular and luteal cellules by increasing the availability of sterol precursor (Grummer and Carroll, 1991). Based on this hypothesis it is expected that progesterone and cholesterol concentration has a positive correlation while the results reported in this case cannot conclude.

Explaining the different results in different experiments, it is stated that the relationship between cholesterol and progesterone can be influenced by fat source, breed, reproductive status and different fat concentration (Lammoglia et al., 1997). Fat supplement will increase absorption of fatty acids from small intestines but cholesterol is essential for this matter. Because while fatty acids are absorbing as chylomicrons or packing like lipoproteins so they could be transferred in lymph or blood (this also occurs in liver). Cholesterol is one of the components of these lipoproteins, so more fatty acids absorbed there is more absorption of cholesterol (Grummer and Carroll, 1991). From total number of 12 experiments about the effects of fat on progesterone levels 9 cases were referred to the increase in progesterone (Ghoreishi, 1998; Burke et al., 1996; Carroll et al., 1990; Estienne et al., 1989; Hawkins et al., 1995; Kuran et al., 1999; Lucy et al., 1993; Spicer et al., 1993; Zamiri et al., 2002). On the other hand, there are three experiments which all show that there is no change in blood progesterone in ruminants regarding to fat sources. These results are all consistent with the present experiment (Sadeghipanah, 2002; Espinoza et al., 1998; Titi and Kridli, 2008). No change in progesterone level, with no improvement in the reproductive characteristics is consistent to this experiment. However, it seems that there is a relation between type of fat and changes in blood parameters and progesterone levels which understanding it requires further studies.

Results in this experiment show that level and combination of Megalac fatty acids on 5.5% of DM for reproduction improvement is not enough, but consumption of 5.2% of Megalac in ewes feed about 2 weeks before birth will improve the lamb weight. Also, consumption of Megalac during mating will increase cholesterol levels, besides it has no effect on blood progesterone levels.

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