

Effect of different Levels of Biosaf Probiotic in Diet of Late Pregnant and Lactating Iranian Zandi Ewes, on Growth Performance and Immune System of their Lambs

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Abstract

A feeding trial was conducted in order to evaluate the effect of different levels of Biosaf probiotic (*Saccharomyces cerevisiae* sc47) in diet of late pregnant and early lactating Zandi ewes on ewe body condition score and growth performance and immune system of their suckling lambs. The experiment was performed on 27 pregnant Iranian Zandi ewes with similar conditions (3-4 years of age, 2-3 parities and at the same time of estrus). According to a completely randomized design, ewes were allocated randomly to one of three treatment (9 ewes/treatment) including: 1-basal diet without probiotic (Control, C) 2-basal diet supplemented with 3 g probiotic per ewe (Low Probiotic, LP) 3-basal diet supplemented with 4.5 g probiotic per ewe (High Probiotic, HP). Basal diet was containing 2.27 MCal/Kg ME and 10.68% crude protein according to NRC 1984. The experiment was conducted 30 days before and 60 days after lambing (suckling period). Lambs live weight was measured at birth and every two weeks interval until Weaning (60 days). Blood samples were collected at first week of birth and weaning and analyzed for blood urea nitrogen (BUN), glucose, globulin, albumin, total protein and white blood cells. Body condition score of ewes measured at beginning of the experiment, after lambing, first month of lactation and weaning. The average daily gain (ADG) of suckling lamb was higher in the HP supplemented group than C and LP groups in all weeks (except week 2), but this superiority reached significant ($P < 0.05$) for lambs at 8 weeks old (168 g/d HP vs. 116 g/d LP and 109 g/d C \pm 12.4). This superiority of ADG of HP suckling lambs happened, despite body condition score of their dams was not significantly difference among treatments ($P > 0.05$). The BUN concentration in HP supplemented groups was lower than C and LP groups indicating improvement in nitrogen efficiency in the rumen. White blood cells were not significantly different among treatment ($P > 0.05$). It is concluded that supplementation of Biosaf probiotic (*Saccharomyces cerevisiae* sc47) to diets of pregnant and lactating of ewes at level 4.5 g/d had positive effects on their lambs growth performance.

Keywords: Probiotic; Suckling lambs; pregnant ewes; Yeast; Growth Performance; Body condition score (BCS); Immune system

Introduction

For many years antibiotics have been used in animal feed as an additive to overcome health problem and improve feed utilization and production efficiency of animals. In spite of several mentioned advantages, the problem of antibiotic resistance of microorganisms was the reason for biotechnology industry to produce other products for replacing antibiotic. Among these additive products was probiotics (bacterial and yeast), which are live microbial feed supplements and particularly during the last 20 years have been used as growth promoters to replace the widely used antibiotics and synthetic chemical feed supplements [1]. In ruminants in this area most research works were carried out with dairy calves or lamb/cattle fattening and less work done with dairy cattle and dairy sheep/goat. Only a few work was carried out to understand the effect of probiotic supplement in diet of pregnant or lactating ruminant particularly sheep on growth performance and immune system of their suckling lambs. In general, in many research pointed out the advantages of using probiotic in ruminant dietary. According to Dawson [2] reported that, yeast culture increased ruminal cellulose digestion, increased microbial growth in the rumen and enhanced microbial protein synthesis and consequently improved feed efficiency and gain. Similarly Williams [3] in his review paper reported that using yeast culture in ruminant diets improve performance. The benefit effects of using yeast culture in performance of growing lamb (Ali [4]; Milewski et al. [5]; Ismaiel et al. [6]; Khalid et al. [7]; Dabiri et al. [8] and goat. As Gado et al.

[9] reported in several works. The published paper of our team clearly pointed out the benefits of probiotic for most works with dairy calves and fattening lambs [8]. So, we do not mention those references here, but focus the effect of probiotic in dairy ruminant, particularly sheep. By using *Saccharomyces cerevisiae* the acid-base balance in dairy cattle improved and resulted in a significantly higher milk production [1,10]. In similar work with sheep, Helal and Abdel-Rahman [11] reported that the increasing milk yield of ewes was an important factor for the production of robust lambs at weaning. Ahmed and Salah [12] and komonna [13] who found that addition yeast culture for diets of ewes during nursing period resulted in improving feed utilization and resulted in better ewe live weight and lamb growth rate. The benefits of effect of probiotic in diet of pregnant ewes on immune system and improved quality of colostrum have shown by Zabek et al. [14] in Turkish. While they did not pointed out to the growth performance and immunity in their suckling lambs, but indirectly it is concluded

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that the greater milk quality and quantity can have a strong positive influence on lamb performance. On contrast, El-Shaer [15] reported that, yeast culture supplementation had no significant effect on final body weight and body gain when sheep fed diet containing (2:1 or 1:2) concentrate: berseem hay ratio with or without 0.25 g yeast culture/10 kg LBW. Rahmani et al. [16] reported higher average daily gain and weaning weight in suckling lambs with creep feed enriched by Yeast *Saccharomyces cerevisiae* ($P<0.05$). Increase blood total protein [15] and glucose concentration [17] and decrease cholesterol [18] and BUN [19] also reported for growing lambs fed dietary containing probiotic, but such results of blood parameters were not found for suckling lambs. The objectives of the present study were to illustrate the effect of Biosaf Probiotic supplementation (Yea-Sacc SC47) in Diet of late pregnant and early lactating of Iranian Zandi ewes on ewe body condition score and growth performance and immune system of their suckling lambs.

Material and Methods

Form a big flock of about 2000 Iranian breeding Zandi sheep in Qum station (120 km in south of Tehran), 27 pregnant ewes with similar conditions (3-4 years of age, and 2-3 parities, that were at the same time of estrus with CIDR and hormone pregnant mare serum gonadotropin (PMSG)) were allocated randomly to one of three treatment diets (9 ewes/treatment) in a completely randomized design. The dietary treatments were included: 1-basal diet without probiotic (Control, C), 2-basal diet supplemented with 3 g probiotic per ewe (Low Probiotic, LP) and 3- basal diet supplemented with 4.5 g probiotic per ewe (High Probiotic, HP). Basal diet was containing 2.27 MCal/Kg ME and 10.68% crude protein according to NRC 1984. Water consumption offered free access. The experimental period was between 30 days before and 60 days after lambing (suckling period). Lambing was occurred within 4 days. Lambs live weight (LW) was measured at birth and during the experiment period (eight weeks) every two weeks interval until Weaning (60 days). Blood samples were collected from jugular vein from 6 lamb in each group before the morning feeding, in two times, at birth and at 8th weeks old of lambs. Samples were analyzed for BUN, Glucose, Globulin, Albumin, Total Protein and White Blood Cells. Body condition score of ewes measured at the beginning of the experiment, after lambing, first month of lactation and weaning. The data were analyzed by GLM procedure for the completely randomized design with three treatments and 9 replicates/treatment using the SAS software (SAS, 1997) and the means were compared using Duncan's Multiple Range test at the level of $P<0.05$.

Results and Discussion

Growth performance

Performance of suckling lambs fed with or without probiotic is shown in Table 1. As shown in Table 1, except in week 2, the average daily gain (ADG) of suckling lamb was higher in the HP supplemented group than C and LP groups in all weeks, but this superiority reached significant ($P<0.05$) for lambs at 8 weeks old (168 g/d HP vs. 116 g/d LP and 109 g/d C \pm 12.4). The lack of significant differences between

treatments groups in suckling lambs in earlier ages in this experiment may contributed to the similar body condition of their dams during pregnancy and lactation (see later part). While the benefit effects of using yeast culture in diets on performance of growing/fattening lambs were recorded in several works (Ali [4]; Milewski et al. [5]; Ismaiel et al. [6]; Khalid et al. [7]; Dabiri et al. [8]), but the results of growing lambs in their experiments are not comparable with the results of suckling lambs were used in this experiment. Also the result reported by Rahmani et al. [16] who have found higher average daily gain and weaning weight in suckling lambs with creep feed enriched by Yeast *Saccharomyces cerevisiae* ($P<0.05$) cannot be a good comparable, because in present experiment the dams of sulking lambs fed probiotic in diets not their lambs. So, as mentioned in introduction few works were carried out to understand the effect of probiotic in pregnant/lactating ewes on their sulking lambs. By using *Saccharomyces cerevisiae* the acid-base balance in dairy cattle improved and resulted in a significantly higher milk production [1]. In similar work with sheep, the benefit effects of probiotic in diet of pregnant ewes on immune system and improved quality of colostrum have shown by Zabeck et al. [14] in Turkish. While they did not pointed out to the growth performance and immunity in their suckling lambs, but indirectly it is concluded that the greater milk quality and quantity can have a strong positive influence on lamb performance. In addition of these indirect agreements, the results of present experiment are in accordance with those reported by Ahmed and Salah [12] and komonna [13] who found that addition yeast culture for diets of ewes during nursing period resulted in improving feed utilization and resulted in better ewe live weight and lamb growth rate. Helal and Abdel-Rahman [11] reported that the increasing milk yield of ewes was an important factor for the production of robust lambs at weaning. Also, El-Ashry et al. [17] and Helal and Abdel-Rahman [11] came to the same conclusion for probiotic or dry yeast supplementation for sheep. On contrast, El-Shaer [14] reported that, yeast culture supplementation had no significant effect on final body weight and body gain when sheep fed diet containing (2:1 or 1:2) concentrate: berseem hay ratio with or without 0.25 g yeast culture/10 kg LBW. In conclusion, the findings of this study revealed that, supplementation Biosaf probiotic (*Saccharomyces cerevisiae* sc47) to diets of ewes at levels 4.5 g/d had positive and beneficial effects on lambs growth performance in older ages.

Blood parameters

Blood constituents of suckling lambs are shown in Table 2. As shown in Table 2, albumin in C group was significantly higher than other group at first week of birth ($P<0.05$), but not at weaning. Plasma blood urea nitrogen (BUN) was significantly ($P<0.10$) lower in the HP supplemented groups than C and LP groups. For other blood parameters including, Glucose, total protein and Globulin, there were not significant differences among treatments ($P>0.05$). The lower BUN concentration in HP supplemented groups than C and LP could be a reason for improvement in nitrogen efficiency in the rumen [7]. This result can be supported by higher growth performance in HP suckling lamb group as explained. In agreement with this result, Dabiri et al. [8] also reported that BUN was lower for the same breed of growing lambs fed dietary containing similar probiotic in higher level. On the other hand, Mukhtar et al. [16] noted that the BUN and creatinine were also non-significant ($P>0.05$) in all treatments groups of growing lambs fed on concentrate with or without Probiotics. According to Mašek et al. [19] also found no difference in blood urea concentration when they studied the influence of live yeast culture (*Saccharomyces cerevisiae*) on milk production and composition, and blood biochemistry of grazing dairy ewes during the milking period. Unlike the results reported by

Week	Treatment			SEM
	Control	Low probiotic	High Probiotic	
2 th	223	213	217	10.45
4 th	183	181	198	7.06
6 th	248	255	271	16.12
8 th	109 ^b	116 ^b	168 ^a	12.42

^{a,b}Value with different Superscripts within a row differ significantly ($p<0.05$).

Table 1: Effect of probiotic supplementation in diet of dams on daily gain performance of their suckling lambs at different weeks (gr/d).

Item	Treatment				Treatment			
	First week of birth			SEM	Weaning			SEM
Control	Low probiotic	High probiotic	Control		Low probiotic	High probiotic		
BUN (mg/dl)	31.93	29.4	26.8	2.08	37.95 ^a	36.70 ^a	31.50 ^b	1.53
Glucose (mg/dl)	86.83	74.33	78	2.77	80.66	77.2	79.83	1.85
Albumin (g/dl)	2.01 ^a	1.81 ^b	1.84 ^b	0.13	3.61	3.53	3.35	0.07
Total protein (g/dl)	6.96	6.6	6.15	0.18	6.7	6.6	6.48	0.09
Globulin (g/dl)	2.88	3.28	2.73	0.14	3.08	3.06	3.13	0.07

^{a,b}Value with different Superscripts within a row differ significantly (p<0.05).

Table 2: Effect of probiotic supplementation in diet of dams on blood parameters concentration of their suckling lambs in 2 times.

Item	First week of birth			SEM	Weaning			SEM
	Control	Low probiotic	High probiotic		Control	Low probiotic	High probiotic	
White blood cell count (/μl)	14716	10400	10280	1411.4	14250	11783	12650	869.5
Neutrophil Segmented (%)	33.16	38.5	35.8	2.09	31.16	34.66	32.83	1.14
Neutrophil Band (%)	0	0.5	0.2	0.13	0.16	0.16	0.16	0.09
Lymphocyte (%)	65.33	60.66	62.6	2.19	66.66	64.5	66.16	1.18
Atypical Lymphocyte (%)	0	0	0	0	0	0	0	0
Monocit (%)	1	0.33	0.8	0.22	1.50 ^a	0.33 ^b	0.83 ^{ab}	0.17
Basophil (%)	0	0	0	0	0	0	0	0
Eosinophil (%)	0.5	0.16	0.6	0.21	0.5	0.33	0	0.1

^{a,b}Value with different Superscripts within a row differ significantly (p<0.05).

Table 3: Effect of probiotic supplementation in diet of dams on white blood cells concentration of their suckling lambs.

Komonna [13], Helal and Abdel-Rahman [11] and Baiomy [20] for sheep during milking period, El-Badawi et al. (1998) found that higher supplementation levels of YC led to higher (P<0.05) plasma urea-N, while total protein content in the blood was stable and comparable between groups in lactating Baladi goats supplemented with YC at 0,1 and 2 g/kg in concentrate feed mixture. The glucose concentration result of this experiment are in agreement with reported result of Ding et al. (2006) who stated that differences in blood glucose concentrations in lambs fed or not fed with the probiotic was not significant (P>0.05). On the other hand, Antunovic et al. [21] reported lower concentration of glucose in lamb (60-day old) supplemented with 0.1% probiotic PDFM. The obtained results for blood Albumin and globulin are in accordance with those reported by El-Shaer [15], Mahrous and Abou-Ammou [22], Komonna [13], Dabiri et al. [8] for sheep and Kholif [23] for goats. They found that YC supplementation did not affect blood Albumin and globulin. However, Khattab et al. [24] with sheep and Shahin et al. [25] with buffalo calves recorded a decrease in blood Albumin and globulin due to YC supplementation. The contradictory in blood parameter results reported by different studies can contribute to several factors e.g. Animal species, physiological conditions, environment and diet and more.

White blood cells as an indicator of immune system

White blood cells concentration and white blood cell count of suckling lambs in different treatment groups are shown in Table 3. Comparison of White blood cell and White blood cells concentration measured at beginning and end of trial showed that there were no significant difference among treatments (P>0.05) for all white cells, except for monocyte, which it was significantly higher in C group than LP group (P<0.05). According to research conducted by Rahman et al. [16], the suckling lambs fed creep-feed containing yeast had no significant effect on the white blood cells. In an experiment with the same breed of growing lambs fed dietary containing same probiotic, Dabiri et al. [18] reported that except for neutrophils and lymphocytes percentages the differences of all other parameters including differential white blood cells between treatments were not significant (P>0.05). Probiotics could lead to increase macrophage activity that stimulates the immune system, which act by increasing the phagocytic ability

of microorganisms. Neutrophils a phagocytosis, which increased the number of band neutrophils in response to probiotic supplements can increase stimulation of macrophage activity [26,27]. Moreover the similar good body condition of ewes as shown in this experiment (see later) reflecting nutritional condition can cause the lack of differences for white blood cells and their concentrates among treatments.

Ewe body condition scores

Body condition scores of ewes fed with or without probiotic are shown in Table 3. About the effect of probiotic on ewes Body condition scores, there were not observed any significant differences between treatments (P>0.05). Body condition of the ewes had no significant effect on live weight. According to research conducted by Gibbal et al. [28] showed that ewes with good body condition more than 3 have better live weight than lean ewes with 2.5. Al-Sabbagh [29] found that Ewes with scores of 3 have better weight in weaning lambs than the ewes with higher score, and lambs had high birth weight in ewes with higher score but it wasn't significant. Data in Table 4 showed that all reproductive traits were similar in the three treatment groups and did not affect by Biosaf probiotic supplementation. In addition, it could be observed that Kilograms of lambs born/ewes lambed was the highest value in group supplemented with 4.5 g/d sc47 (4.71) followed by 3 g/d sc47 supplemented group (4.67) and then control group (4.66). Also, kilograms of lambs weaned/ewes lambed followed the same pattern. These results are agreement with obtained by komonna [13] and Helal and Abdel-Rahman [11] who reported that supplementation of yeast to ewe diets increased birth weight and weight gain of their offspring. Saccharomyces cerevisiae reported to balance the energy and the acid-base metabolism in dairy cattle resulted in a significantly higher milk production [1]. Similarly, Masek et al. [30] and Helal and Abdel-Rahman [11] came to the same conclusion for sheep and reported increasing milk yield of ewes is an important factor for the production of robust lambs at weaning. According to Ismaiel et al. [6] reported that yeast culture increased average daily gain of lambs. Dawson [31] showed that, yeast or yeast culture are rich source of vitamins, enzymes and other important nutrients and co-factors which make them attractive as digestive enhancers as a basic source of nutrients [32-34].

Times	Treatment			SEM
	Control	Low probiotic	High probiotic	
Last month of pregnancy	2.83	3.16	2.91	0.09
After lambing	3.08	3.58	3.41	0.11
First month of lactation	3.58	3.58	3.41	0.13
Weaning	3.5	3.5	3.33	0.14

^{a,b}Value with different Superscripts within a row differ significantly ($p < 0.05$).

Table 4: Effect of probiotic supplementation on body condition score of ewes during late Pregnancy and 3 times during lactation.

Conclusions

In conclusion, the findings of this study (improved ADG and lower BUN in suckling lambs) revealed that, supplementation of Biosaf probiotic (*Saccharomyces cerevisiae* sc47) to diets of pregnant and lactating of ewes at level 4.5 g/d had positive and beneficial effects on their lambs growth performance in older ages. The lack of differences in immune indicators may contribute to the similar body condition of their dams in different treatments groups. More studies with pregnant ewes under different feeding levels and body conditions are necessary to clarify the effects of live yeast cell supplementation to ewe diets on their suckling lambs.

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