

Effect of Rumen Undegradable Protein (RUP) on Colostrum Quality and Growth of Lori Bakhtiari Lambs

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Abstract: This experiment was conducted to determine the effect of Rumen Undegradable Protein (RUP) on colostrum quality and lambs growth. 60 lori bakhtiari ewes with 2 and 3 parities which were in last month of gestation were randomly selected and divided into 3 groups. Ration, was include of the control diet (without RUP), 40% RUP of total crude protein and diet contain of 60% RUP of total crude protein. Each group of ewes randomly used one of the diets for 4 weeks before lambing. In this experiment fish meal was fed as a source of rumen undegradable protein. Completely randomized design with 3 treatments and 20 replicates per treatment was used. Results showed that birth weight of lambs was increased with increasing levels of RUP, but it was not statistically significant. Increasing of RUP effectively improved weaning weight of lambs and SNF (Solid Not Fat) percentage of primary colostrum ($p < 0.05$). It also was effective on percentage of protein and first phase colostrum lactose ($P < 0.01$). Impact of increasing RUP from 40 to 60 percent on weight and colostrum composition of the second phase (12 hr after lambing) and milk components of first, second and third phase (SNF percentage, protein percentage, fat percentage and lactose percentage) was not significant. With increasing in rumen undegradable protein level from 40 to 60 percent, concentration of immunoglobulins M, A, G of colostrum boosted ($p < 0.01$). The concentration of immunoglobulins M, A, G of ewes serum also significantly ($p < 0.01$) improved. Results showed that the use of undegradable protein to 60 percent of total dietary protein can promote colostrum quality, birth weight and weaning weight of lambs.

Abbreviations: RUP, Rumen Undegradable Protein

Key word: Rumen Undegradabl Protein • Colostrum • Ewes • Immunoglobulin

INTRODUCTION

There are several years during which crude protein is used to determine the protein requirement of livestock. But new methods of feedstuffs evaluation show that, individually crude protein is not enough to investigate the impact of protein on animal's growth and performance. Furthermore, it doesn't estimate the protein requirement of ruminants accurately [1]. To improve the protein utilization in ruminant feed, protein should be divided to its components. It means that we should be able to distinguish, how much protein is degraded in the rumen and how much is bypassed from the rumen and absorbed in the gut. To find the optimum ability of ruminant with high production (meat and wool), rumen degradable

protein does not meet their needs [1]. So rumen undegradable protein is necessary, to provide essential amino acids for their growth [2]. Amount of digested and absorbed protein in the small intestine is an important factor in the body and wool growth [3, 4]. Protein supplements which have proper levels of rumen undegradable protein provide growth limiting amino acids like lysine and methionine. Fish meal is a valuable protein supplement which provides these two amino acids as well as RUP [5]. Fish meal contains approximately has 10% lipid including a significant amount of long chain n-3 polyunsaturated fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) [6]. Nutrition in late pregnancy is a key factor influencing lamb birth weight, mammary development and colostrums

production. Fifty percent of foetal growth takes place during the final 4 weeks of pregnancy alongside major redistribution of the ewe's body reserves to maintain udder development and colostrum production. An adequate intake of quality colostrum within 24 h of birth is essential for lamb survival. Newborn lambs are hypo-immunocompetent and have limited reserves of brown adipose tissue (BAT) and hepatic glycogen for heat production. Ingestion of pre-formed cloistral immunoglobulin's (IgG) and metabolic substrates is essential to provide protection against early post-natal infection and prevent starvation or hypothermia [7]. When the supply of microbial protein is limited by energy intake, detrimental effects on colostrums production can be ameliorated by increasing the supply of amino acids through the provision of rumen undegradable protein [8].

The purpose of this experiment was to investigate the impact of RUP consumption during late gestation on protein production and increasing of immunoglobulin concentration to promote reproduction performance of ruminants.

MATERIALS AND METHODS

In this experiment, 60 Lori-Bakhtiari ewes out of 250 with 2 and 3 parities which were in last month of gestation were randomly selected from a herd, located near Esfahan-Shahrekord road and used in this experiment. All ewes were healthy and in a proper body condition. They were selected in mating season and synchronized with progesterone soaked sponge. After 14 days sponge was

removed and all ewes were injected by 1 dose PMSG hormone (permangole, 500 IU or 2.5 ml). Once ear tags were punched, all ewes were divided in 3 different treatments (20 ewes each of them) with 5 replications and then determined by blue, red and white colors. After that, ewes were transported to 3 different ranches for 1 month and each ranch was divided with 5 cages. Also, ration, was include of the control diet (without RUP), 40% RUP of total crude protein and diet contain of 60% RUP of total crude protein. Each group of ewes randomly used one of the diets for 4 weeks before lambing. The components of experiment diets showed in table 1. Feed and water were adlibitium and ration was fed 3 times a day. Management was the same for all sheep. Ration ingredients were contain of alfalfa hay, barley grain, heat treated soybean, beet pulp, rapeseed meal, salt, calcium carbonate, phosphorus supplement and fish meal which substituted rapeseed meal in the experiment. After a month of feed consumption and lambing season initiation, feed consumption and body weight gain were recorded. After parturition ear tags were used for identification. Body weight after birth, lambs genes, kind of birth and parturitions condition was recorded. Daily colostrum production was recorded quickly after birth. At first colostrum was milked from one teat and weighted and another one prepared for lamb to eat. Weighting of lambs was done before and after colostrum consumption. Ewes were milked two times a day. A colostrum sample was taken from the first and next 12 hour milking and collected in a 50 ml plate and transported to the lab in the refrigerator for the next analysis.

Table 1: The components of experiment diet

Food	Deit 1Control	Deit 2 (40% RUP of total crude protein)	Deit 3 (60% RUP of total crude protein)
Alfalfa hay	40.00	40.00	40.00
Barley grain	35.50	33.00	30.00
Rapeseed meal	15.50	5.50	0.00
Beet pulp	6.50	12.00	17.00
Heat treated soybean	0.00	4.50	6.00
Salt	0.50	0.50	0.50
Calcium carbonate	0.50	0.50	0.50
Phosphorus	1.50	1.50	1.50
Fish meal	0.00	2.50	4.50
Energy metabolism(mcal/kg)	2.41	2.43	2.43
Crude protein (%)	16.10	16.10	16.10
Rumen undegradable protein (%)	30.70	33.90	36.20
Rumen degradable protein (%)	66.80	63.60	61.30
Neutral detergent fiber (%)	32.50	32.40	32.70
Acid detergent fiber (%)	20.60	20.60	20.90
Calcium (%)	1.24	1.36	1.47
Phosphor (%)	0.47	0.47	0.47

The most important factors which measured are contain of 1-IgG immunoglobulin 2-IgM immunoglobulin 3-IgA immunoglobulin 4-protein 5-Lactose 6-SNF (Solid Not Fat) 7-fat. Colostrum samples were also diluted and dilution was taken into account for analysis. Blood samples were taken after lambing, sent to lab and analyzed with electrophoresis method [9]. Milk production were recorded 3 times during 3 month, in a way that each month during the first week milk production were recorded, morning and evening samples mixed then a uniform sample collected in the special 50 ml plates and analyzed for protein, Lactose, SNF, fat in the lab. To measure the milk components such as protein, Lactose, SNF, fat milkoscan S50 was used. This experiment was conducted in the form of complete randomized design with 3 treatments and 20 replications. The collected data were analyzed by SAS statistical software [10] and comparison of means was analyzed by t-test.

RESULTS

Comparison of means (\pm SD) of birth weight, lambs weaning weight (90 days), first and second phase of colostrum weight and first, second and third phase of milk weight, are shown in table 2. Birth weight of lambs was not significantly different. With increasing of RUP, birth weight and lambs weaning weight were increased. Lambs weaning weight was highest in treatment 3 (60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60% RUP of total crude protein) was not significantly different from treatment 2 (40% RUP of total crude protein). But it was different from treatment 1 (control) ($p < 0.05$). Difference lambs weaning weight between treatments 1 (control) and 2 (40% RUP of total crude protein) was not significant. Amount of colostrum in first and second phase and also amount of milk in all

3 phases were 2ot significantly different. With increasing of RUP, amount of colostrum in first and second phase and also amount of milk in all 3 phases were increased.

Comparisons of means (\pm SD) related to first and second phase of colostrum components, are shown in table 3. SNF percentages of primary colostrum highest in diet contain 60% RUP of total crude protein and lowest in Control. Treatment 3 (60% RUP of total crude protein) was not significantly different from treatment 2 (40% RUP of total crude protein). But it was different from treatment 1(control) ($p < 0.05$). Difference SNF percentage of primary colostrum between treatments 1(control) and 2(40% RUP of total crude protein) was not significant. Protein percentage of primary colostrum highest in treatment 3 (60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3 (60% RUP of total crude protein) was not significantly different from treatment 2 (40% RUP of total crude protein). But it was different from treatment 1 (control) ($p < 0.01$). Difference protein percentage of primary colostrum between treatments 1 (control) and 2 (40% RUP of total crude protein) was not significant. Therefore we can conclude that increasing of RUP to the 60 percent of crude protein, elevated SNF percentage and protein percentage of primary colostrum. Fat percentages of primary colostrum were not significantly different. Lactose percentage of primary colostrum highest in treatment 1 (Control) and lowest in treatment 3 (60% RUP of total crude protein). Treatment 1 (Control) was different from treatment 2(40% RUP of total crude protein) ($p < 0.01$). Also it was different from treatment 3(60% RUP of total crude protein) ($p < 0.01$). Therefore we can conclude that Lactose percentage of primary colostrum was decreased, with increasing of RUP. Also weight, percentage of SNF, Fat and second phase colostrums protein (12 hours after lambing) were not significantly different and with increasing of RUP, this components were increased.

Table 2: Comparison of means (\pm SD) of birth weight, lambs weaning weight (90 days), first and second phase of colostrum weight and first, second and third phase of milk weight

Item	Treatment		
	(1) (Control)	(2) (40% RUP of total crude protein)	(3) (60% RUP of total crude protein)
Birth weight (kg)	4.21 \pm 0.09 ^a	4.20 \pm 0.09 ^a	4.28 \pm 0.09 ^a
Weaning weight (kg)	32.09 \pm 0.30 ^b	32.67 \pm 0.31 ^{ab}	32.69 \pm 0.30 ^a
First phase colostrum weight (g)	539.23 \pm 26.08 ^a	559.34 \pm 27.25 ^a	577.18 \pm 26.08 ^a
Second phase colostrum weight(g)	652.20 \pm 35.52 ^a	694.59 \pm 37.09 ^a	699.71 \pm 35.52 ^a
Frist phase milk weight (g)	706.21 \pm 35.31 ^a	744.00 \pm 36.87 ^a	754.05 \pm 35.30 ^a
Second phase milk weight (g)	758.57 \pm 39.99 ^a	836.80 \pm 41.76 ^a	862.72 \pm 39.99 ^a
third phase milk weight (g)	758.57 \pm 39.99 ^a	836.80 \pm 41.76 ^a	862.72 \pm 39.99 ^a

First and second phase's colostrum components

Table 3: comparison of means (\pm SD) related to first and second phase of colostrum components

Item	Treatment		
	(1) (Control)	(2) (40% RUP of total crude protein)	(3) (60% RUP of total crude protein)
SNF percentage of primary colostrum	25.03 \pm 0.23 ^b	25.44 \pm 0.23 ^{ab}	25.85 \pm 0.23 ^a
Fat percentage of primary colostrum	6.45 \pm 0.14 ^a	6.49 \pm 0.15 ^a	6.67 \pm 0.14 ^a
protein percentage of primary colostrum	13.62 \pm 0.18 ^b	14.19 \pm 0.19 ^{ab}	14.52 \pm 0.18 ^a
Lactose percentage of primary colostrum	3.53 \pm 0.05 ^a	3.43 \pm 0.06 ^a	3.30 \pm 0.05 ^b
Colostrum SNF percentage of second phase	21.61 \pm 0.19 ^a	21.68 \pm 0.20 ^a	21.75 \pm 0.19 ^a
Colostrum Fat percentage of second phase	6.19 \pm 0.14 ^a	6.16 \pm 0.14 ^a	6.19 \pm 0.14 ^a
Colostrum protein percentage of second phase	10.93 \pm 0.19 ^a	11.01 \pm 0.19 ^a	11.17 \pm 0.19 ^a
Colostrum Lactose percentage of second phase	3.23 \pm 0.05 ^a	3.21 \pm 0.05 ^{ab}	3.11 \pm 0.05 ^b

First, second and third phases of milk components

Table 4: comparison of means (\pm SD) related to first, second and third phase of milk components

Item	Treatment		
	(1) (Control)	(2) (40% RUP of total crude protein)	(3) (60% RUP of total crude protein)
Milk SNF percentage of first phase	19.87 \pm 0.24 ^a	19.93 \pm 0.25 ^a	20.05 \pm 0.24 ^a
Milk Fat percentage of first phase	5.87 \pm 0.15 ^a	5.85 \pm 0.15 ^a	5.88 \pm 0.15 ^a
Milk protein percentage of first phase	8.25 \pm 0.18 ^a	8.20 \pm 0.19 ^a	8.25 \pm 0.18 ^a
Milk Lactose percentage of first phase	4.39 \pm 0.09 ^a	4.38 \pm 0.10 ^a	4.53 \pm 0.09 ^a
Milk SNF percentage of second phase	19.07 \pm 0.19 ^a	19.30 \pm 0. 20 ^a	19.28 \pm 0.19 ^a
Milk Fat percentage of second phase	6.10 \pm 0.13 ^a	6.18 \pm 0.13 ^a	6.25 \pm 0.13 ^a
Milk protein percentage of second phase	7.46 \pm 0.19 ^a	7.61 \pm 0.19 ^a	7.56 \pm 0.19 ^a
Milk Lactose percentage of second phase	4.14 \pm 0.12 ^a	4.11 \pm 0.13 ^a	4.13 \pm 0.12 ^a
Milk SNF percentage of third phase	19.07 \pm 0.19 ^a	19.30 \pm 0.20 ^a	19.28 \pm 0.19 ^a
Milk Fat percentage of third phase	6.10 \pm 0.13 ^a	6.18 \pm 0.13 ^a	6.25 \pm 0.13 ^a
Milk protein percentage of third phase	7.47 \pm 0.19 ^a	7.61 \pm 0.19 ^a	7.56 \pm 0.19 ^a
Milk Lactose percentage of third phase	4.14 \pm 0.12 ^a	4.13 \pm 0.13 ^a	4.13 \pm 0.12 ^a

immunoglobulins of colostrum and ewes blood serum

Table 5: comparison of means (\pm SD) related to colostrum immunoglobulins and ewe blood serum

Item	Treatment		
	(1) (Control)	(2) (40% RUP of total crude protein)	(3) (60% RUP of total crude protein)
immunoglobulin G of colostrum (mg/dl)	4160.83 \pm 141.83 ^b	4776.11 \pm 148.09 ^a	5150.83 \pm 141.83 ^a
immunoglobulin A of colostrum(mg/dl)	422.61 \pm 15.09 ^b	478.79 \pm 15.77 ^a	493.61 \pm 15.09 ^a
immunoglobulin M of colostrum(mg/dl)	627.66 \pm 16.84 ^c	676.41 \pm 17.59 ^b	743.16 \pm 16.85 ^a
immunoglobulin M of ewe blood serum(mg/dl)	1756.23 \pm 43.39 ^b	1889.91 \pm 45.31 ^a	1993.73 \pm 43.39 ^a
immunoglobulin M of ewe blood serum(mg/dl)	32.78 \pm 1.93 ^b	34.73 \pm 2.00 ^{ab}	42.78 \pm 1.93 ^a
immunoglobulin M of ewe blood serum(mg/dl)	183.82 \pm 5.13 ^c	194.44 \pm 5.35 ^b	215.32 \pm 5.13 ^a

Colostrum lactose percentage of second phase (12 hours after lambing) highest in treatment 1 (Control) and lowest in treatment 3(60% RUP of total crude protein). Treatment 1(Control) was not different from treatment 2(40% RUP of total crude protein).but it was different from treatment 3(60% RUP of total crude protein) ($p < 0.01$). Therefore Colostrum Lactose percentage of second phase was decreased, with increasing of RUP.

Comparison of means (\pm SD) related to first, second and third phase of milk components, are shown in table 4. Percentage of SNF, Fat, protein and first phase milk Lactose (First week of first month after lambing), milk of second phase (First week of second

month after lambing) and milk of third phase (First week of third month after lambing) were not significantly different. With increasing of RUP, milk SNF percentage of first phase, milk fat percentage of second phase and milk fat percentage of third phase were increased. Percentage of fat and first phase milk protein, Percentage of SNF and second phase Milk protein and percentage of SNF and third phase first milk protein were not affected by RUP consecutively but when the ration contain of 40 and 60 percent RUP were fed, mentioned factors decreased compared to control group. Milk lactose of second and third phase was decreased by increasing of RUP.

Comparison of means (\pm SD) related to colostrum immunoglobulins and ewe blood serum, are shown in table 5. Amount of immunoglobulin G of colostrums was highest in treatment 3(60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60% RUP of total crude protein) was different from treatment 1 (control) ($p < 0.01$), but it was not significantly different from treatment 2(40% RUP of total crude protein). Amount of immunoglobulin A of colostrum was highest in treatment 3 (60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60% RUP of total crude protein) was different from treatment 1 (control) ($p < 0.01$), but it was not significantly different from treatment 2 (40% RUP of total crude protein). Amount of immunoglobulin M of colostrums was highest in treatment 3 (60% RUP of total crude protein) and lowest in treatment 1(Control). Treatment 3(60% RUP of total crude protein) was different from treatment 2 (40% RUP of total crude protein) ($p < 0.01$). Also it was different from treatment 1(control) ($p < 0.01$). Treatment 1(control) was different from treatment 2 (40% RUP of total crude protein) ($p < 0.01$). Amount of immunoglobulin G of ewes blood serum highest in treatment 3 (60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60% RUP of total crude protein) was different from treatment 1 (control) ($p < 0.01$). But it was not significantly different from treatment 2(40% RUP of total crude protein). Amount of immunoglobulin A of ewes blood serum highest in treatment 3(60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60% RUP of total crude protein) was different from treatment 1 (control) ($p < 0.01$), but it was not significantly different from treatment 2 (40% RUP of total crude protein). Also treatment 2 (40% RUP of total crude protein) was not significantly different from treatment 1 (control). Amount of immunoglobulin M of ewes blood serum was highest in treatment 3(60% RUP of total crude protein) and lowest in treatment 1 (Control). Treatment 3(60 RUP of total crude protein) was different from treatment 2 (40% RUP of total crude protein) ($p < 0.01$). Also it was different from treatment 1 (control) ($p < 0.01$). Treatment 1(control) was different from treatment 2 (40% RUP of total crude protein) ($p < 0.01$). So we can conclude that with increasing of RUP, colostrum immunoglobulin and blood serum of ewes were increased.

DISCUSSION

Birth weight of lambs was increased with increasing levels of RUP which was not consistent with the results of others [7, 11, 12]. This is a probable reason for

inconicantly that lamb birth weight is more sensitive to dietary protein level than degradability parse [13, 14].

Weaning weight of lambs was increased with increasing levels of RUP. At birth (0 d), lambs born to the fish meal supplemented ewes had greater plasma content of EPA, DHA and total VL_n-3-PUFA. However, the contents of these FA were further increased for the lambs that received colostrum and milk from mothers supplemented with fish meal. Sheep colostrums and milk can be enriched with DHA by dietary fish meal supplementation during late gestation and lactation led to increased lambs weaning weight [15].

With increasing levels of RUP, First and second phase of colostrum percentage of SNF, protein and first and second phase colostrum fat were increased. Hall *et al.* [12] observed significant increase in colostrums yield following protein supplementation of ewes at pasture. This response is likely to have arisen from the increased nutrient intake of supplemented ewes, as observed by [16]. These results in contrast to those observed [7, 11]. These results suggested that the level of dietary protein, rather than its degradability, may be of greater importance for ensuring high levels of colostrum production in ewes [7].

With increasing of levels RUP, amount of milk in all 3 phases was increased. These results are in agreement with several other studies [17, 21]. fish meal led to increase in ratio of grams of AA N:megajoules of metabolizable energy (ME), a further increase in absorbed AA N increased the volume of milk produced by supplying gluconeogenic substrates for lactose synthesis [22]. Another reason was the benefit of supplemental RUP the increased flow of AA to the small intestine. Reported that rumen-protected met and Lys increased milk yield in sheep. Fishmeal, an RUP source high in Met and Lys, increased milk yield in sheep by 17% compared with untreated soybean meal [21]. In other experiment, the highest milk yield was better explained by increased DMI rather than by enhanced body lipid mobilization [23]. Also Fish meal increased duodenal supply of limiting a necessary for higher production. Indicated that the excess protein may have been supplying gluconeogenic substrates for lactose synthesis or for meeting other energy requirements [24]. Increasing in Milk yield is contrast to several other results [25, 29]. Forage-to-concentrate ratio, stage of lactation, solubility of FM protein and diet composition are variables that may be related to the variation in milk response to fish meal supplementation [26]. The lack of yield response to RUP has been attributed to the inability of the protein supplement to increase the total quantity or improve the quality of protein flowing to the small intestine [28, 29].

With increasing of levels RUP, milk fat percentage in all 3 phases was increased. These results are in agreement with several other studies [21,30-33]. However, fish meal feeding increased milk fat production likely in response to an increase in the ratio of absorbed AA N: ME [30]. Also not reduce milk fat percentage compared with control group, indicating that fish meal supplementation might be less potent to fiber utilizing rumen microbes, especially cellulolytic bacteria [15]. This increase not agreement with other results [34, 27]. Given the lipid content of fish meal, ruminal fiber digestion may be decreased by the presence of polyunsaturated fatty acids (PUFA) in fish meal [38]. Fish meal contains approximately 10% residual lipid and significant amounts of long-chain n-3 PUFA [6]. Suggested that high concentrations of high molecular weight polyunsaturated fatty acids in fish oil may alter microbial flora in the rumen, resulting in decreased acetate to propionate ratio or a decrease in uptake of plasma fatty acids by the mammary gland [28]. Also, this depression of milk fat can be attributed to a negative effect on the acetate and propionate concentrations, alterations of fat metabolism posttruminally, or inhibition of ruminal microbial fatty acid synthesis [24]. Two conditions seem necessary for milk fat depression: presence of rumen substrate in the form of dietary unsaturated fatty acids and an altered rumen environment that leads to incomplete biohydrogenation and production of various substrates and conjugated linoleic acid isomers [39].

Milk protein percentage was not affected by RUP consecutively but when the ration contain 40 and 60 percent RUP were fed, mentioned factors increased compared to control group. These results are in agreement with other results [6, 40]. suggested that ruminant fed diets of alfalfa silage showed greater increases in milk protein percentage when fed fish meal than did ruminant fed other forages, perhaps because such diets generally provide less flow of essential fatty acids to the duodenum [6]. Feeding fish meal highly resistant to ruminal degradation will improve the total supply and amino acid profile and, thus, increase the supply of limiting amino acids for milk protein synthesis [40]. The intestinal supply of AA that usually limit milk protein synthesis might have increased when fish meal was fed, which might explain the increased protein content in milk from ruminant fed the fish meal [41].

By increase of levels RUP, second and third phase milk lactose percentage was decreased compared control group that this result is agreement with other results [30, 33] suggested that, even for fish meal diets, energy supply was adequate, but AA N supply limited any further increase in FCM production [30].

CONCLUSION

In conclusion, supplementation of RUP had positive effects on colostrum quality and growth of loribakhtiar lambs. According to these observations, the use of undegradable protein to 60 percent of total dietary protein can promote colostrum quality, birth weight and weaning weight of lambs.

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