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Dietary polymer-coated urea enhances the goats lactational performance, excretion of microbial purine derivatives and blood metabolites in the semi-arid zone of Iran

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ABSTRACT. This study aimed to determine the effect of using different sources of nitrogen to supply part of degradable intake protein needs in lactating goat performance and its effect on excretion of microbial purine derivatives and blood metabolites. Thirty-two lactating Saanen goats (body weight 38.85 ± 2.14 kg and 1979 ± 0.25 g day^{-1} milk yield) were used in a one-way ANOVA completely randomized design. Goats were assigned to the following treatments for a 10-wk experimental period: 1) Control (canola meal as a nitrogen source); 2) Urea (0.5% urea); 3) Optigen (0.55% Optigen- Alltech. Inc., Lexington, KY) and 4) Polymer-Coated Urea (PCU- international patent number: A01K5/00, 0.7% PCU) based on dry mater intake. Non-protein nitrogen groups had a comparative effect ($p > 0.05$) between control and other treatments on milk composition, microbial protein synthesis and they affected on blood factors including urea, cholesterol, and ALT. Dry matter intake decreased ($p > 0.05$) in PCU, Optigen, Urea than Control goats. Synthesis of microbial protein in PCU goats was 22.5 g day^{-1} and it was greater ($p > 0.05$) than other treatments. Plasma cholesterol was increased in PCU and Optigen, whereas urea concentration was increased in Urea and Control goats. Milk production was higher in PCU than Urea and Control. Feed conversion ratio was improved ($p > 0.05$) in PCU and Optigen goats *versus* other treatments. This study demonstrated that polymer-coated urea can be utilized as a nitrogen source and improve goats milk performance.

Keywords: goat; microbial synthesis; milk yield; optigen; urea.

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Introduction

Nutrition has a major effect on animal production systems including the goat raising farms and has a remarkable effect on the cost of productions (Romero-Huelva, Ramírez-Fenosa, Planelles-González, García-Casado, & Molina-Alcaide, 2017). Goat feed sources are usually pasture and crop by-products. Until 50 year ago, just a few and limited research had been published on goat nutrition and this was mostly because they were assumed to be pets, not livestock, and destroyed the environment, and therefore did not deserve scientific treatment, this attitude resulted in the lack of research for solving goat farmer's questions (Amills, Capote, & Tosser-Klopp, 2017). But now it is known that goat farming has some benefits, one benefit is that goats can be milked once a day without decreasing milk production (Mazinani & Rude, 2020). Milk of goats persistency will be up to 2 to 3 years even without kidding, which means goats can provide winter milk without needing seasonal breeding (Baker & Miller, 2019).

As feed cost is one of the main costs of goat raising systems, using alternative products such as urea can be an economic solution, but it was associated with ammonia toxicity because urea rapidly hydrolyzed to ammonia in the rumen under the action of urease (Stocker et al., 2013). If the releasing rate of urea is controlled, rumen microorganisms can utilize ammonia then, microbial bodies will digest and absorb post-rumen. Using biuret and protecting urea with different coated materials have been developed to achieve this goal. Due to now, several slow-release urea products have been developed and tested for this purpose (Spanghero, Nikulina, & Mason, 2018). The results of these research are contradictory, and rate of nitrogen release is different depends on affecting parameters such as rumen

situation, feed processing and forage to concentration ratio. Theoretically, the urea breakdown rate should be increased to shorten the degradation (Alipour et al., 2020). Polymer-coated urea (PCU) is one of the newest coated urea products that coated with oil materials. This product showed positive effects in slowing rate of urea degradation urea compare with other expensive coated-urea products in previous studies (Mazinani, Naserian, Mesgaran, & Valizadeh, 2018).

Considering these benefits for raising goats and also effects of nutrition management on the cost of end terminal products, the objective of this study was to use different sources of nitrogen for supplying part of the protein needs of goats and determining goat response and performance while consuming different nitrogen sources.

Materials and Methods

Animals, treatments and management

Thirty-two lactating Saanen goats were used in a 10-wk lactation trial, confinement each goat in individual metabolism cage. As it is shown in Figure 1, goats were randomly assigned to each of four different nitrogen (N) source diets (8 goats per treatment) and each animal was fed individually on a daily basis. Treatments were: 1) control (canola meal as a N source); 2) Urea (0.5% urea); 3) Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen) and 4) Polymer-coated urea (0.7% PCU) (international patent number: A01K5/00, inventor: Mitra Mazinani).

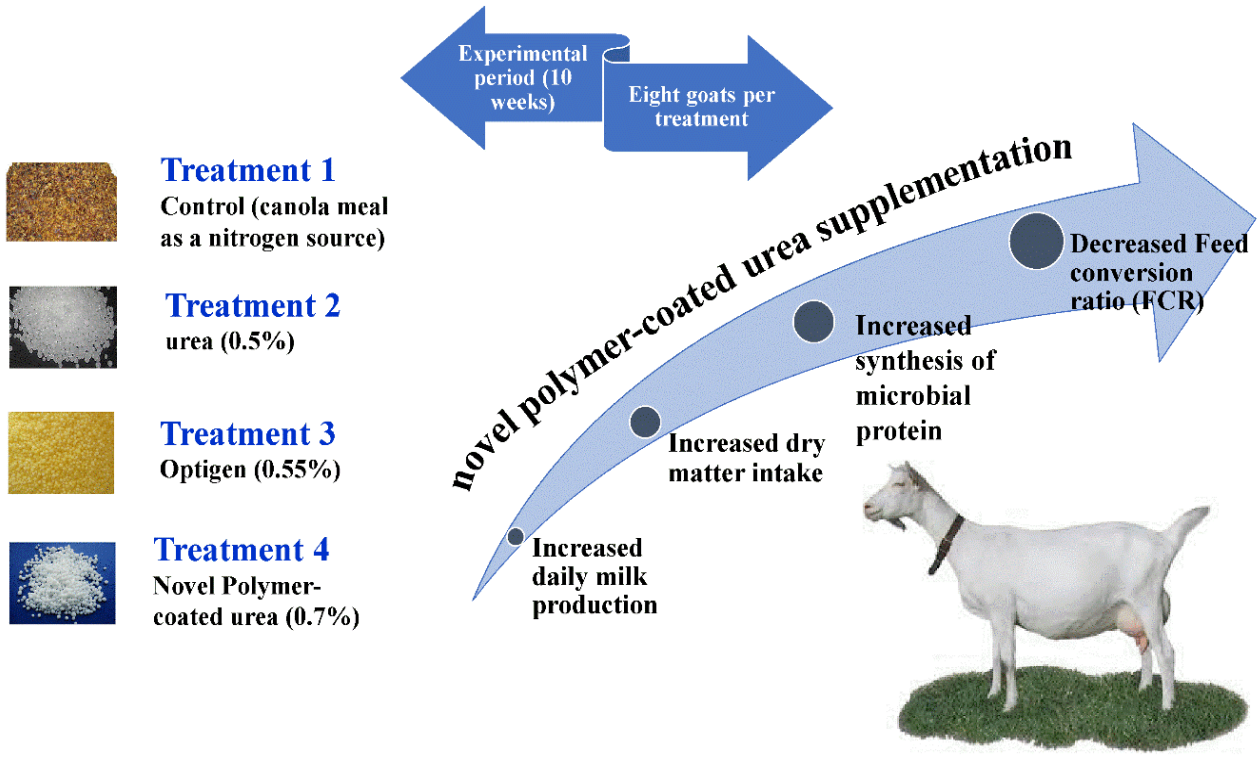

Figure 1. Graphical abstract of experimental treatments.

In Table 1, diets were formulated to satisfy the nutrient requirements of Saanen goats using Small Ruminants NRC version 1.9.4468 (Tedeschi, Callaway, Muir, & Anderson, 2011). Goat's average weight was 38.85 ± 2.14 kg, and 1979 ± 0.25 g d^{-1} milk yield, and fed *ad libitum* twice daily (08:00, 15:00 h) isonitrogenous total mixed ration (15% crude protein) and they were housed in individual pens (Animal Care Committee Protocol: 37756).

Feed intake Nutrient digestibility

Daily amounts of feed distributed, and feed refusal were weighed. Fecal and feed refusal samples were taken weekly. Apparent nutrient digestibility was calculated by measuring the difference between the consumed feed and feces as a percentage of goats feed intake and considering lignin as an internal marker (Cochran, Adams, Wallace, & Galyean, 1986).

Table 1. Diet composition and chemical analysis of different dietary treatments (% of dry matter)[1].

Diet composition (% DM)	Control	Urea	Optigen	PCU
Alfalfa hay	40	40	40	40
Wheat straw	5	5	5	5
Corn grain	27.5	27.5	27.5	27.5
Canola meal	17.5	11	10.95	10.8
Wheat grain	8	14	14	14
Calcium carbonate	0.6	0.6	0.6	0.6
Salt	0.5	0.5	0.5	0.5
Urea	0	0.5	0	0
PCU	0	0	0	0.7
Optigen	0	0	0.55	0
Min-Vit Mix[2]	0.9	0.9	0.9	0.9
Chemical analysis (% DM)				
Dry matter	90.40	90.32	90.31	90.33
Organic matter	80.20	80.46	80.43	80.62
Crude protein	15.54	15.20	15.05	14.78
Neutral detergent fibre	37.00	39.90	39.88	39.85
Acid detergent fibre	21.40	21.00	21.00	20.95

[1]Control (canola meal as dietary N source); Urea (0.5% urea); Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen), and Polymer-Coated Urea (PCU, 0.7% - patent number: A01K5/00). [2]Each kg contained: Vit A, 500000IU; Vit D3, 100000 IU; Vit E, 100mg; Ca, 190000mg; P, 90000mg; Na, 50000mg; Mg, 19000mg; Fe, 3000mg; Cu, 300mg; Mn, 2000mg; Zn, 3000mg; Co, 100mg; I 100mg; Se, 1mg; Antioxidant (B.H.T) 3000mg.

Measurements and analysis

Feed, feed refusal and feces were analyzed for crud protein (CP) by using copper catalyst and steam distillation into boric acid according the Kjeldahl method (method 973.18) and for ash (method 942.05), acid detergent fiber (ADF, method 973.18) as initial marker according AOAC methods (ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTIS [AOAC], 2005). Neutral detergent fiber (NDF) was estimated according to method of Soest, Robertson and Lewis (1991). Feeds refusal and feces were collected and dried for 24 h at 105°C in oven to measure dry matter content (method 934.01) and then samples were grounded with 1-mm screen of Wiley mill. Complete fecal collection of each goat was done in collection periods (once each week and every day in collection week).

Microbial Nitrogen synthesis

Urine samples were taken according to Chen, Mejia, Kyle and Ørskov (1995) to determine microbial crude protein. Urine was collected hourly with using a fraction collector. In order to pH decrease under 3, an initial amount of H_2SO_4 was added in 30 ml bottles before collecting. If the final pH of samples was greater than 3, more H_2SO_4 solution was added to adjust it. In practice, urinary excretion of purine derivatives is an indicator of microbial protein and it was measured as the sum of allantoin, uric acid, xanthine and hypoxanthine using a method based on the AutoAnalyzer (Chen & Gomes, 1992).

Blood metabolites

The blood samples were taken fortnightly 2 hours after first feeding in morning, from the jugular vein of goats by using vacutainer tubes containing heparin and centrifuged at 2,500 RPM for 20 min. Plasma was harvested and frozen at -20°C. Total protein, albumin, cholesterol, urea, triglyceride, ALT and AST were measured by AutoAnalyzer (Biosystems A 15; 08030 Barcelona, Spain- Pars azma kits).

Milk production and composition

Goats were milked twice daily with machine (GEA portable model, USA) at 7 am and 3 pm during feeding time, and daily milk yield was recorded throughout the trial period. Milk samples were collected weekly. In the sampling week (last 7 days), 50 ml milk of each goat was thoroughly homogenized, and milk samples were analyzed for protein, fat, lactose and total solids-non-fat content using an automatic Milkoscan 605 analyser (Foss Electric, Hillerød, Denmark, AOAC International, 2005). Milk energy (MJ kg^{-1}) was estimated using fat, lactose and CP content of the milk for each goat (Tyrrell & Reid, 1965).

Statistical Analysis

Data were analyzed by one-way ANOVA analysis of variance using the general linear models (GLM) procedure of SAS 9.1. Nitrogen treatments were considered the only sources of variation. The significance of

differences between control and experimental treatments was estimated by using one-way ANOVA, with Duncan's post-hoc test, and alpha level of $p < 0.05$ was used to assess the significance among means. The statistical models used in this study were as follows:

$Y_{ij} = \mu + T_i + e_{ij}$

where: Y*ij* = observed response, μ = overall mean, T_i = treatment effect (I = 1 to 4), E_{ijk} = residual error

Results

Total diet consumption in polymer-coated urea (PCU) and Optigen were more ($p > 0.05$) than Control goats. However, consumption of crude protein, NDF and ADF were not affected by treatments (Table 2).

Table 2. Effect of the polymer-coated urea product and Optigen on dry matter intake of goats (g head^{-1} day^{-1})[1].

	Control	Urea	Optigen	PCU	SEM[2]	p-value[3]
DM intake	1985.01[c]	2105.52[ab]	2157.77[a]	2047.84[a]	23.419	0.020
Crude protein	304.27	325.71	333.70	308.19	12.085	0.845
Organic matter	1785.38[ab]	1894.24[ab]	1938.61[a]	1833.52[ab]	21.361	0.023
Acid detergent fiber	419.01	449.99	473.01	430.03	12.977	0.533
Neutral detergent fiber	724.5	854.9	899.7	816.5	19.545	0.129

[1]Control (canola meal as dietary N source); Urea (0.5% urea); Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen), and Polymer-Coated Urea (PCU, 0.7% - patent number: A01K5/00).[2]SEM: standard error of mean. [3]Means within same row with different superscripts differ ($p < 0.05$).

Synthesis of microbial protein was not affected ($p > 0.05$) by the treatments, however, PCU goats synthesized more microbial protein than other groups, that indicates adding non-protein nitrogen increased synthesis of microbial protein (Table 3). Cholesterol was increased in PCU and Optigen goats than Control. Blood urea concentration was increased in Urea and Control goats. The PCU and Urea treatments increased the plasma ALP concentration (Table 4).

Table 3. Effect of the polymer-coated urea product and Optigen on microbial nitrogen synthesis[1].

Items	Control	Urea	Optigen	PCU	SEM[4]	P-value[5]
Microbial nitrogen synthesis (g day^{-1})	20.95	17.99	18.93	22.49	0.772	0.150
PD[2] absorption (mmol day^{-1})	28.81	24.74	26.04	30.94	1.014	0.107
PD excretion (mmol day^{-1})	26.20	22.79	23.88	27.99	0.875	0.132
Allantoin excretion (mmol day^{-1})	22.27	19.37	20.29	23.79	0.814	0.222
Uric acid excretion (mmol day^{-1})	3.93	3.42	3.58	4.20	0.187	0.508
DOMR[3] (mmol day^{-1})	0.65	0.56	0.59	0.70	0.039	0.658

[1]Control (canola meal as dietary N source); Urea (0.5% urea); Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen), and Polymer-Coated Urea (PCU, 0.7% - patent number: A01K5/00). [2]PD: purine derivatives. [3]DOMR: digestible organic matter apparently digested in the rumen. [4]SEM: standard error of mean. [5]Means within same row with different superscripts differ ($p < 0.05$).

Table 4. Effect of the polymer-coated urea product and Optigen on blood metabolites of goats[1].

Item	Control	Urea	Optigen	PCU	SEM[5]	P-value[6]
Total protein (g L^{-1})	85.84	83.07	87.54	87.79	1.892	0.850
Albumin (g L^{-1})	37.66	36.97	35.49	39.49	1.305	0.804
Cholesterol (mg dL^{-1})	50.68[ab]	44.65[b]	55.95[ab]	57.75[a]	1.936	0.038
TG[2] (mg dL^{-1})	29.37	31.32	32.30	25.62	1.366	0.356
Urea (mg dL^{-1})	29.23[a]	29.47[a]	25.60[b]	27.40[ab]	0.557	0.014
ALT[3] (U L^{-1})	21.15[c]	24.55[a]	22.59[ab]	23.48[a]	0.485	0.050
AST[4] (U L^{-1})	92.68	96.76	95.39	104.92	2.504	0.386

[1]Control (canola meal as dietary N source); Urea (0.5% urea); Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen), and Polymer-Coated Urea (PCU, 0.7% - patent number: A01K5/00). [2]TG: three glycerides. [3]ALT: alanine aminotransferase. [4]AST: aspartate aminotransferase. [5]SEM: standard error of mean. [6]Means within same row with different superscripts differ ($p < 0.05$).

Milk production was changed by treatments, and there was a significant difference between Urea and Control goats. Milk production increased ($p > 0.001$) in Optigen and PCU goats *versus* other treatments. Feed conversion ratio was better ($p > 0.05$) in PCU and Optigen goats than other treatments. Milk composition (fat, lactose, protein, solids, and total solids non-fat) was not affected ($p > 0.05$) by treatments (Table 5).

Table 5. Effect of the Polymer-Coated Urea product and Optigen on milk yield and contents of goats[1].

Item	Control	Urea	Optigen	PCU	SEM[2]	P-value[3]
Milk yield (g day^{-1})	1857.13^{b}	2100.89^{ab}	2335.4^{a}	2317.8^{a}	62.466	< 0.001
Feed conversion ratio	1.17^{a}	1.02^{b}	0.9^{bc}	0.86^{c}	0.044	0.021
Fat (g day^{-1})	4.41	4.2	3.68	3.66	0.215	0.134
Total solid non-fat (g day^{-1})	8.91	8.55	8.073	8.37	0.440	0.946
Lactose (%)	4.63	4.71	4.45	4.61	0.224	0.986
Protein (%)	3.09	3.145	2.968	3.075	0.071	0.885
Energy (MJ kg^{-1})	67.67	74.54	71.36	77.63	2.183	0.464

[1]Control (canola meal as dietary N source); Urea (0.5% urea); Optigen (Alltech. Inc., Lexington, KY) (0.55% Optigen), and Polymer-Coated Urea (PCU, 0.7% - patent number: A01K5/00). [2]SEM: standard error of mean. [3]Means within same row with different superscripts differ ($p < 0.05$).

Discussion

Feed intake and digestibility

It was observed due to supplementing slow-release urea in a high-forage diets, dry matter intake decreased (Neal, Eun, Young, Mjoun, & Hall, 2014). Cherdthong, Wanapat and Wachirapakorn (2011) found that the urea and slow-release urea (urea-calcium sulfate and urea-calcium chloride) did not influence dry matter intake. However, Pinos-Rodríguez, Peña, González-Muñoz, Bárcena and Salem (2010) observed that consuming slow-release urea did not affect dry matter intake. The results of this research are consistent with the results of the present exp. The mechanism of supplementing slow-release urea on feed intake is difficult to explain but can be related to forage content and synchronization with non-starch carbohydrates of diet and slow-release urea percent in diet. Feed intake and digestibility are intimately related to each other and may affect N availability in rumen (Köster et al., 1996). Some studies reported positive influences on digestibility and feed intake when ruminants consuming slow-release urea instead to feed grade urea. Cherdthong et al. (2011) conducted a study on supplementing calcium-urea in diet of dairy cows that fed rice straw and results showed calcium-urea mixture increased organic matter intake and digestibility, compared to feed grade urea. Owens, Lusby, Mizwicki, and Forero (1980) compared effect of supplementing feed grade urea and lipid coated slow-release urea on steers fed cottonseed hulls and found slow-release urea increased cottonseed hull intake but digestibility did not change. In contrast, Taylor-Edwards et al. (2009) and Galo, Emanuele, Sniffen, White and Knapp (2003) fed polymer-coated urea to dairy cows using corn silage base diet and did not observe any improvement in dry matter intake or digestibility. However, the effect of urea and non-protein nitrogen sources on digestibility depends on their concentration, physical structure and adaptation period. Increased digestibility of Optigen and coated urea could be due to these treatments supplied enough nutrients for microorganisms in the rumen.

Microbial nitrogen synthesis

The role of supplementing ruminant diets with non-protein nitrogen is supplying ammonia for ruminal bacteria as a nitrogen source for synthesizing amino acids. Although, high releasing of ammonia can exceed the nitrogen utilization capacity of bacteria and reduce productive performance (Calomeni et al., 2015). It has been reported, oils can defaunation the rumen (Puniya, Singh, & Kamra, 2015), so maybe due to this fact, existence of paraffin (oil) as a coated material in the polymer-coated urea which increased microbial protein synthesis by eliminating protozoa. The synthesis of microbial nitrogen for 'coated urea' was greater than other treatments and this difference was also observed in other parameters of microbial nitrogen synthesis. Optigen and control treatments had the greater microbial synthesis, respectively, which indicates using non-protein nitrogen may increase the synthesis of microbial protein, although this difference was not significant. The purpose of any nutritional strategy that is designed to improve microbial protein synthesis is accomplished by improving the final production of the animals or increase efficiency of animal production. Cherdthong et al. (2011) reported an increase in total milk yield by dairy cows fed urea-calcium.

Blood metabolites

Mazinani et al. (2018) reported that supplementing urea can elevate blood urea concentration more than canola meal and it is due to rapid hydrolysis of urea to ammonia in the rumen .excess ammonia is absorbed into the blood from the rumen wall and then taken to the rumen where urea is usually detoxified. part of this urea can recycle by the rumen while the excess amount is excreted in the urine (Lapierre & Lobley, 2001).

Other authors have reported that slow-release urea reduced blood urea concentration compared with feed grade urea (Cherdthong et al., 2011; Taylor-Edwards et al., 2009), that agrees with present results.

Milk production and composition

Xin, Schaefer, Liu, Axe and Meng (2010) showed different slow-release urea sources did not affect milk fat. Cameron, Klusmeyer, Lynch, Clark, and Nelson (1991) found milk production was increased by supplementing urea, whereas protein and fat concentrations were not altered. In another study, Supplementing slow-release urea increased milk protein (Tye, Yang, Eun, Young, & Hall, 2017). Dennis, Unruh-Snyder, Neary and Nennich (2012) reported that goats' milk composition was not affected by dietary protein concentration, and this agrees with the present results. Xin et al. (2010) also observed no change in milk protein with a slow-release urea treatment. It can be concluded that feed efficiency was improved with the addition of urea and other non-protein nitrogen sources and increased milk efficiency. According result, coated urea and Optigen had better feed conversion ratio grade compared with control or feed grade urea. In recent study slow-release urea increased milk production but no impact on milk composition. This results was in contract with Santos and Huber (1996) reported that replacing part of soybean meal by Optigen or feed grade urea had no impact on milk yield but agreed with Inostroza, Shaver, Cabrera and Tricárico (2010) reported of increasing milk production when Optigen partially replaced with feed grade urea.

Conclusion

This study revealed the benefits of supplementing Saanen goat diet with polymer-coated urea. Adding polymer-coated urea in the diet positively influenced dry matter intake, milk yield, and feed conversion ratio. The Microbial nitrogen synthesis in the polymer-coated urea goats showed a tendency to higher compared to other treatments. Based on economical, this experiment demonstrated that the polymer-coated urea can be utilized as a non-protein nitrogen supplement and Supply part of rumen digestible protein while reducing feed and finial goat production costs. In future studies, it is suggested that the effect of polymer-coated urea be investigated in large scale animals such as dairy cows and feedlot steers.

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