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The impact of saponin and tannin on performance, blood constituents, and carcass fatty acids in Baluchi lambs

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Abstract The aim of this experiment was to study the effect of pure saponin and tannic acid on feed intake, weight gain, immune metabolites, blood lipids, and fatty acid profiles of lamb carcass. In this experiment, 18 lambs with an average weight of 17±1.5 kg were used in a completely randomized design (3 treatments with 6 replications) for 60 days. Treatments consisted of a basal diet consisting of forage (30%) and concentrates (70%), and diets containing saponin (150 mg/kg dry matter) or tannic acid (15 g/kg dry matter). Dry matter intake, weight gain, feed conversion ratio, packed cell volume and blood concentrations of hemoglobin, glucose, urea nitrogen, total proteins, and triglycerides were not affected by the experimental treatments. Concentration of saturated fatty acids and unsaturated fatty acids with a double bond did not show a significant difference between the experimental treatments, but the sum of unsaturated fatty acids with several double bonds was highest in the tannic acid treatment ($P<0.05$).

Keywords: lamb, saponin, tannic acid, carcass composition

Introduction

Cardiovascular diseases are said to be a leading cause of death in many countries. Elevated concentrations of cholesterol or low density lipoprotein (LDL) in the blood have been identified as the most important risk factors for heart disease. Inheritance, high blood pressure, obesity, inactivity, smoking, and poor diet are the main factors predisposing people to these diseases (Naghshi et al., 2020).

Meanwhile, the composition of the human diet plays an important role in preventing disease and controlling the concentration of lipids and lipoproteins in the blood. Reducing cholesterol and saturated fat consumption is the primary goal of diet therapy in reducing the incidence of cardiovascular disease. Meat consumption increases blood cholesterol in humans and some animal species due to their high levels of cholesterol and saturated fats (Campbell, 2017). The role of ruminants as the most important food supply organisms for humans is quite palpable. Therefore, in the current situation, it is tried to lead the pattern of rumen fermentation and microbial ecosystem of the animal to the production of useful animal products through genetic manipulation and the use of special compounds. Due to the special conditions in their digestive system, ruminants have the ability to ferment many compounds found in their feeds, depending on the input of food, have the ability to produce a variety of substances that can have a direct impact on animal health and function (Ozturk and Gursel, 2021).

The presence of anti-nutrients in animal feed can have

a significant impact on the performance and health of the animal. Anti-nutrients are divided into 4 groups: A) substances such as protease inhibitors, saponins, tannins, and lectins that reduce the efficiency of use and digestibility of protein B) inhibitors of minerals such as oxalate, phytate, and gossypol C) anti-vitamin substances such as coumarin and D) other anti-nutritional substances such as mycotoxins, mimosine, cyanogen, nitrate, alkaloids, and isoflavones (Yacout, 2016). Considerable research is currently being done to evaluate the potential of anti-nutritional compounds to modify ruminal fermentation. Saponins and tannins are among the most important of these compounds and play important roles in the ruminal fermentation pattern. On the other hand, tannins and saponins have inhibitory effects on the activity and population of ruminal microorganisms that are responsible for biohydrogenation (Yanza et al., 2021). The last stage of biohydrogenation of unsaturated fatty acids is inhibited by saponins and tannins, which leads to increased accumulation of vaccenic acid and decreased stearic acid (Khiaosa et al., 2009). Alfalfa, soybeans, and sugar beet pulp, which are the mainstays of livestock and poultry feed, are rich in saponins. In addition, due to the shortage of animal feed in developing countries, farmers are forced to use unconventional ingredients in animal diets, which are usually rich in tannins and saponins. Therefore, in both developed and developing countries, tannins and saponins are present in most oral compounds (Popova and Mihaylova, 2019).

There have been many studies on the use of saponins and tannins in the process of fermentation activities, but information in this field is not complete, requiring more research on the effects of saponin and tannin on ruminal activity. Therefore, the aim of this study was to investigate the effect of recommended levels of saponin and tannic acid on feed intake, nutrient digestibility, blood attributes, and fatty acid profile of lamb carcass.

Materials and methods

Animals and the diets

The experiment was conducted at the Livestock and Poultry Research Center of the Faculty of Agriculture, Ferdowsi University of Mashhad. Baluchi lambs were kept in individual metabolic cages (1.5×1.5 m), equipped with individual manger and water trough. The animals had free access to water during the day and night. This experiment was performed from the first of May to the end of June in a barn at an average temperature of 20 °C and the mean humidity of 48% under natural lighting conditions.

In this experiment, 18 lambs with an average weight of 17±1.5 kg were used in a completely randomized design (3 treatments and 6 replications per treatment) for 60 days. Before starting the experiment, the lambs were examined for leg and hoof conditions, infectious and metabolic diseases by a veterinarian. The animals were vaccinated against Foot-and-Mouth, Smallpox, Brucellosis, Charbon, Liver gangrene, and Enterotoxemia (Rooyan Darou Company), and treated for worms (Albendazole 2.5%, Tolide Darouhai Dami Iran Company).

Experimental treatments included one level of pure saponin (Loba Chemie PVT. LTD. Mumbai. India) at 150 mg/kg dry matter and tannic acid (Merck, Germany) at 15 g/kg dry matter; added to the basal diet containing forage (30%) and concentrate (70%). The basal diet was formulated for maintenance and 200 g daily weight gain. The diets were fed as total mixed ration (TMR) and offered to the lambs in two equal meals at 08:00 and 20:00 hours. Ingredients and chemical composition of the basal diet are shown in Table 1.

Table 1. Ingredients and chemical composition of the basal diet

Ingredients	Percentage in dry matter
Barley silage	30
Corn grains	13
Barley grains	24.25
Rapeseed meal	10.5
Soybean meal	7
Wheat bran	10.5
Vitamin and mineral supplements	0.7
Salt	0.35
Ground lime stone	0.7
Sunflower oil	3
Chemical composition	
Crude protein	15.22
Ether extract	6.3
Neutral detergent fiber (NDF)	34.7
Non fibrous carbohydrate (NFC)	38.9
Metabolizable energy (Mcal/kg DM)	2.52

Concentration of the dry matter, ether extract, ash, and crude protein in feed and fecal samples were measured by the AOAC methods (AOAC, 2002), and acid detergent fiber (ADF) and neutral detergent fiber (NDF) according to Van Soest et al. (1991).

For calculation of the average daily gain, the lambs were weighed at the beginning and the end of the experiment at 8 a.m. after a 12 hour- fast during which water was freely available. Daily feed consumption was determined as the total feed offered minus orts.

Blood sampling

Blood samples were taken from the jugular vein on the last day of the experiment, before morning feeding. Sub-samples were kept at room temperature for 30 minutes followed by refrigeration for 10 minutes. Coagulated samples were centrifuged for 10 minutes at 3000 rpm and serum samples were transferred by syringe to special plastic containers and stored at -20 °C. A 5 mL-subsample of blood was transferred into EDTA-coated CTA tubes, gently shaken, and immediately transferred to the clinical pathology laboratory of the Faculty of Veterinary Medicine, Ferdowsi University of Mashhad for

determination of white blood cells, hemoglobin, whole blood protein and packed cell volume (PCV) in whole blood using a cell counter (Model CellTac-α; Nihonkohden, MEK, 6450K, Japan). Serum glucose, cholesterol, triglycerides, and urea nitrogen were measured using the Biosystem Co. kits and Automatic analyzer (Auto Analyzer A15, Biosystem S.A. Barcelona, Spain).

Ruminal fluid sampling

Ruminal fluid was collected on the last day of the experiment, three hours after morning feeding, using an esophageal tube and with the help of a suction pump. Ruminal fluid was pH was immediately measured by pH meter (Metrohm Company, Model 691). The fluid was strained through four-layer mesh cloth, and 10 mL fluid was mixed with 10 mL 0.2 N hydrochloric acid (Merck Co.) in special plastic containers and immediately stored in a freezer at -20 °C.

To measure ammonia nitrogen, a 5 mL sample of the mixture was transferred into a Tecator manual Kjeltec digestion flask and titrated using sodium tetraborate and 0.01 N hydrochloric acid solution. Concentration of ammonia nitrogen was calculated based on the volume of titrated acid and standard values (amount of 1 mL of ammonium sulfate + 2.5 mL of 0.2 normal hydrochloric acid) (Naserian, 1996).

Meat sampling

The profile of intramuscular fatty acids was measured at the end of the experimental period, using three lambs per treatment. The lambs were anesthetized by a veterinarian, and a sample of *Longissimus thoracis* (approximately, 8 g) was prepared by biopsy.

A. Lipid extraction: About 40 grams of minced meat and 80 mL methanol were mixed gently for two minutes. Then, 80 mL of methanol were added and mixed for two minutes, followed by addition of 80 mL chloroform and mixing for two minutes, after which, water was added according to the water content of the sample. The mixture was filtered through vacuum paper number one and then transferred to a separating funnel. After phase separation, chloroform was collected and the solvent was evaporated using a rotary evaporator (Folch et al., 1957).

B. saponification: To one gram of the extract, six mL methanol were added and shaken gently to obtain a uniform mixture. A solution of 4 mL of potassium hydroxide in methanol was refluxed for five minutes under nitrogen gas and five minutes without nitrogen gas for 20 minutes at 90 °C.

C. Methylation: Three mL boron trifluoride were added to 50 mL of oil and mixed using a magnetic stirrer for 30 minutes. Then three mL of distilled water were added to the solution, and one hour was allowed for phase separation. The organic phase of the sample was extracted three times with two mL hexane using a separating funnel. The mixed organic phase and half a gram of anhydrous magnesium sulfate were added to remove the excess moisture and then filtered through a Whatman grade paper and injected into a gas chromatograph.

D. Gas Chromatography-Mass Spectrometry (MS-GC) Separation of methyl esters: This was done by using a gas chromatograph with a TurboMass Auto system XL mass spectrometer connected to a flame ionization detector (FID) and a computer with Total Chrom Navegator (Aligent) software 7890B). Samples were injected 20% without gaps in the column of polyethylene glycol (RTX-2330), RESTEK 0.25 mm, (Elite WAX) 105 m. Injector and FID temperatures were maintained at 230 °C. The temperature programming of the system was as follows: 180 ° C for three minutes followed by an increase to 250 °C, this temperature was maintained for 15 minutes (Hashemipour et al., 2013).

Statistical analysis

The data were analyzed by Proc GLM of SAS 9.1 software in a completely randomized design using the following statistical model:

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

Y_{ij}: individual observation, μ: mean of the total population, T_i: effect of treatment, Σ_{ij}: random effect of error. The Tukey's test at 5% error level was used to compare the treatment means.

Results

Feed consumption, weight gain, and feed conversion ratio

Dry matter intake, weight gain, and feed conversion ratio were not affected by experimental treatments (Table 2; P>0.05).

Table 2. The effect of dietary saponin and tannic acid on weight gain, dry matter intake, and feed conversion ratio in Baluchi lambs

	Experimental diets			SEM	P- value
	Control	Saponin	Tannin		
Initial weight (kg)	16.75	17.01	16.93	0.87	0.97
Final weight (kg)	26.08	25.04	25.40	1.03	0.77
Average daily weight gain (g/day)	155.44	133.75	141.06	7.97	0.18
Average dry matter intake (g/day)	765.40	738.22	716.43	18.12	0.19
Feed conversion ratio	4.96	5.58	5.23	0.31	0.41

Ruminal pH and ammonia nitrogen and blood constituents

Ruminal fermentation parameters (pH and ammonia nitrogen) and most blood constituents were not different between the experimental treatments (Table 3; P>0.05). Total blood protein level tended to be higher in control lambs (P=0.09). Total white blood cells in the control treatment were lower than in the saponin and tannin groups (P<0.05).

Table 3. The effect of dietary saponin and tannic acid on ruminal fermentation and blood constituents in Baluchi lambs

Metabolite	Experimental diets			SEM	P- value
	Control	Saponin	Tannin		
Ruminal parameters					
Ammonia nitrogen (mg/dL)	18.93	17.08	17.53	0.65	0.14
pH	6.31	6.27	6.43	0.11	0.57
Blood constituents					
Glucose (mg/dL)	73.56	72.14	75.94	1.37	0.18
Blood urea nitrogen (mg/dL)	36.12	34.68	34.19	1.28	0.55
Total protein (mg/dL)	50.75	47.98	48.42	0.88	0.09
Triglycerides (mg/dL)	15.93	17.28	16.57	0.43	0.12
Cholesterol (mg/dL)	57.27	55.66	55.21	1.79	0.60
Packed cell volume (PCV, %)	31.50	31.00	29.00	1.38	0.44
Total protein (g/dL)	5.35	5.35	5.38	0.17	0.99
Hemoglobin (g/dL)	11.73	11.40	11.21	0.18	0.18
Total white blood cells (per µL)	10812^{c}	13413^{b}	15700^{a}	540.41	0.0004

[a,b] Within rows, means with common superscript do not differ (P>0.05)

Profile of Longissimus thoracis fatty acids

There were significant differences in the concentration of individual fatty acids; however, the overall concentrations of the saturated and unsaturated fatty acids, and their ratios, were not influenced by tannin or saponin treatment (Table 4). There was no significant difference in the concentrations of C14:0, C16:0, and C18:0 fatty acids between the experimental treatments (Table 4; P>0.05). The C17:0 concentration in tannin treatment was reduced compared to the control and saponin treatment (P<0.05). The concentration of mono-unsaturated fatty acids (MUFA) (C16:1, C17:1, and C18:1) did not show a significant difference between treatments. The concentration of polyunsaturated fatty acids (PUFA) was highest in the lambs feeding on the tannic acid diet (P<0.05).

Table 4. The effect of dietary saponin and tannic acid on fatty acid profile[1] of the *Longissimus thoracis* in Baluchi lambs

Fatty acid	Experimental diets			SEM	P- value
	Control	Saponin	Tannin		
C14:0	1.40	1.37	1.20	0.17	0.70
C16:0	22.61	21.17	21.55	1.41	0.97
C16:1	3.93	3.97	3.62	0.26	0.62
C17:0	3.97^{a}	3.38^{ab}	2.9^{c}	0.26	0.06
C17:1	1.74	1.85	1.97	0.25	0.82
C18:0	13.50	12.33	11.57	0.57	0.13
C18:1	48.43	50.38	50.49	1.49	0.58
C18:2 n-6	3.93^{ab}	3.83^{b}	4.95^{a}	0.30	0.07
C18:3 n-3	0.41^{b}	0.51^{ab}	0.62^{a}	0.03	0.01
C 20:4 n-6	0.68^{a}	0.35^{b}	0.73^{a}	0.03	0.0001
C20:5 n-3 (EPA)	0.21^{b}	0.48^{a}	0.21^{b}	0.01	0.0001
C22:6 n-3 (DHA)	0.17^{b}	0.37^{a}	0.19^{b}	0.01	0.0003
Saturated fatty acids	40.48	38.25	37.22	1.34	0.29
Unsaturated fatty acids with one double bond	54.10	56.20	56.07	1.58	0.60
Polyunsaturated	5.41^{b}	3.54^{b}	6.70^{a}	0.31	0.04
Total unsaturated	59.51	61.74	62.77	1.61	0.40
Ratio of saturated to unsaturated fatty acids	0.68	0.62	0.59	0.04	0.26

[1] Weight percentage of total fatty acids
[a,b] Within rows, means with common superscript(s) do not differ (P>0.05).

Discussion

Feed consumption, weight gain, and feed conversion ratio

Hervas et al. (2003) reported that the addition of quebracho tannin to sheep diets at levels of 0.5, 1.5, and 3 g/kg body weight did not affect feed intake up to 1.5 g/kg, but at higher concentration, it was significantly reduced.

In experiments on dairy cows, Benchaar et al. (2008) reported that the use of concentrated tannins had no effect on the digestibility of dry matter, organic matter, crude protein, NDF, and ADF. They observed higher growth, less parasitic excretion, improved reproductive performance and wool production in lambs fed Birdsfoot trefoil (*Lotus corniculatus*). One of the reasons for improving reproductive performance and wool production can be an increase in metabolizable protein due to the binding of protein and tannins.

The results of the effect of saponin on feed intake have been contradictory. Some studies have reported a relatively small reduction (2-6%) in feed intake (Lovett et al., 2006; Benchaar et al., 2008). Holtshausen et al. (2009) reported that yucca and quillaja saponins reduced food intake. Lovett et al. (2006) reported that consuming 25 and 50 mg of saponins per day reduced feed intake. Adding 250 mg/kg yucca saponin to the mixed diet (45% forage, 50% rolled barley, and 5% soybean meal) to fattened calves diet (Hussain and Cheeke, 1995) and adding 30 mg/kg saponin to sheep diet (in a diet containing 50% forage and 50% concentrate) had no effect on growth performance (Seliwinski et al., 2002).

Saponin changes the function of ruminants by protozoan reduction, although this difference in function, also depends on the diet and saponin structure. Reducing protozoa may also reduce fiber digestion in the rumen. It is almost universally accepted that protozoan depletion improves the performance of ruminants, especially in low-protein diets. The effect of reducing protozoa depends on the balance between the energy and protein requirement of the animal and the energy and protein supplied by the feed. Protozoan depletion of animals with high protein requirements and diets with real protein levels and low energy is beneficial. Therefore, the effect of saponin depends on the type of diet (Eugene et al., 2004).

Blood constituents

Except for total white blood cells, other blood constituents were not significantly affected by feeding saponin or tannins. Nasri et al. (2011) reported that feeding 0, 30, 60 and 90 mg of saponin per kg dry matter intake to lambs reduced blood glucose levels significantly. Ben Salem et al. (2005) showed that goat diets containing tannins did not cause a significant change in blood glucose and protein concentrations compared to controls. However, concentration of blood urea nitrogen decreased significantly with increasing tannin in the diet. Blood glucose depends on the state of energy balance and the type of diet (Silanikove et al., 2006).

Blood total protein concentration tended to decrease by feeding saponin and tannins in the present experiment. Several studies showed reduction in blood protein level by adding tannins to the diet (Vasta et al., 2009; Mousa, 2011). Formation of a complex between - tannins and proteins removes the protein from the reach of ruminal microbes, and sometimes this complex prevents the post-ruminal digestion and absorption of proteins.

Data concerning the effects of diets containing unsaturated fatty acids and tannins on blood lipid metabolites (triglycerides, cholesterol, HDL, and LDL) are very limited. Mousa (2011), studying the addition of acacia to sheep diets, showed that the level of blood cholesterol in diets containing tannins was not significantly different from the control group.

According to Vasta et al. (2009), tannins affected the composition of saturated, unsaturated, and vaccenic fatty acids in the ruminal fluid and tissue lipids, but not in blood. This was explained by the fact that the ratio of some fatty acids (SFA, MUFA, and PUFA) in blood plasma is different from that in the ruminal fluid and adipose tissue. This obscures the effects of tannins on the percentage of these fatty acids in the blood relative to the rumen and adipose tissue. According to the report of Vasta et al. (2009), measuring the composition of blood fatty acids alone may lead to erroneous interpretation of the results regarding the effects of tannins on ruminal biohydrogenation and lipid metabolism in the blood. It is generally concluded that the concentration of both fatty acids and lipid transporters in the blood should be measured to examine blood lipid metabolites in order to interpret the effect of tannins on ruminal biohydrogenation and lipid metabolism in the blood.

We found no effects of treatments on blood cholesterol level. Saponin and bile acids can form large mixed micelles which increase the excretion of bile acids, thus accelerating cholesterol metabolism in the liver and reducing serum cholesterol (Matsuura, 2001). Nasri et al. (2011) reported that feeding 0, 30, 60 and 90 mg of saponin per kg dry matter intake to lambs had no effect on blood cholesterol and urea concentrations.

Bhatta et al. (2004) reported that the addition of concentrated tannins to the diet caused a significant decrease in hemoglobin concentration. The toxicity of oak leaves and acute disease of sheep and cattle is due to the presence of high levels of hydrolysable tannins. Poisoning is due to the absorption of tannins and the increase of phenols in the blood, which the liver is responsible for detoxifying. Secretion of liver enzymes may increase when high doses of tannins need to be detoxified.

Both saponin and tannic acid caused an increase in the number of white blood cells in the current experiment. Saponin causes anemia and decreases red blood cell count by increasing plasma membrane permeability and decreasing smooth muscle activity. Baumann et al. (2000) reported that saponin damage to the fatty layers of the human erythrocyte membrane is irreversible. Subcutaneous injection of *Panax notoginseng* saponin significantly prevented platelet aggregation and adhesion in rats exposed to cerebrovascular occlusion. The antispasmodic properties of *Panax notoginseng* saponin are exerted on

brain cells, possibly by altering the structure of membrane proteins (by increasing their fluidity, which leads to altered protein function). Platelet membrane glycoproteins play a key role in platelet binding to vessel walls, platelet aggregation, and thrombosis. Seliwinski et al. (2002) reported that feeding at 0.1 g/kg in the diet had no effect on hematocrit or hemoglobin concentration. Hematocrit count and aminotransferase and glutamate dehydrogenase activity were not affected by feeding *Sapindus saponaria* fruits.

Fermentation parameters

Neither ruminal pH nor ammonia nitrogen concentration was affected by feeding saponin or tannins in the present study. Bhatta et al. (2004) using two levels of tannin (12.3% and 21.12% of dry matter in the diet) did not observe a significant difference in the ruminal pH between the sheep fed with tannin and the control group. Also, Yildiz et al. (2005) using oak leaves in the diet of lambs did not observe a change in ruminal pH compared to the control. The pH of the rumen depends on feeding time and production of volatile fatty acids (Silanikove et al, 2006). Studies have shown that tannins reduce the concentration of volatile fatty acids in the rumen and therefore can affect ruminal pH (Silanikove et al., 2006). However, it has also been reported that the rate of effective degradation and degradability of proteins could decrease in the rumen of tannin-fed animals leading to a decrease in ammonia concentration (Bhatta et al., 2004). Nasri et al. (2011) reported that feeding 0, 30, 60 and 90 mg of saponin per kg of dry matter intake to lambs had no effect on ruminal ammonia nitrogen concentration. Alipanahi et al. (2019) reported a significant decrease in ruminal fluid ammonia concentration by adding oak fruit (containing hydrolyzable tannin) to the diet of lactating goats. Saponins can bind to ammonia to prevent excessive ammonia build-up in the rumen, release it when the rumen ammonia concentration decreases, and help build microbial protein. However, saponins can act as mediators when NH_4 is sufficiently and continuously available. Changes in ruminal ammonia levels are affected by two processes, i.e., the rate of protein breakdown in the rumen, and the uptake of ruminal ammonia for microbial protein synthesis (Hussain and Cheeke, 1995).

Data on the effect of saponin on ruminal pH are contradictory. Nasri et al. (2011) reported no significant difference in ruminal pH between treatments receiving different levels of saponin and the control group. Benchaar et al. (2008) stated that adding 60 g of yucca extract per day to the diet of lactating cows did not cause a significant change in the ruminal pH. Lila et al. (2005) showed that feeding 0, 0.5, and 1% sarsaponin in the diet of fattening calves significantly reduced the ruminal pH.

Profile of Longissimus thoracis fatty acids

When the supply of unsaturated fatty acids to the rumen is high or the biohydrogenation process is incomplete, the unsaturated fatty acids are transferred to the lower parts of the gastrointestinal tract and absorbed in the intestine, and deposited in milk and meat (Dhiman et al., 2000).

Two fatty acids, C16:1 and C18:1, are formed in muscles both through food and through the conversion of saturated fatty acids C16:0 and C18:0 by the enzyme delta-9-desaturase. While sic-9 C14:1 is made only in muscles by the enzyme delta-9-desaturase. Therefore, the rate of (C14:0 1 C14:1 cis-9) sic-9 / C14:1 is an indicator for assessing the activity of delta-9-desaturase enzyme in lamb muscles (Palmquist et al., 2004). In the study by Vasta et al. (2009), sic-9 C14:1 content was higher in tannin-containing treatments, indicating that tannin was effective in the expression of delta-9-desaturase. The inability of saponin in affecting the synthesis of fatty acids with one or more double bonds may be due to the habituation of rumen microbes to saponin or the breakdown of saponin by lamb saliva (Teferedegne, 2000).

Tannins are said to be able to decrease saturated fatty acids by reducing bacterial proliferation and inhibiting biohydrogenation. In an experiment conducted by Priolo et al. (2005) with *Hedysarium coronarium* containing 18 g/kg DM tannins, the levels of CLA, C20:5 n-3 (EPA) and MUFA in mutton were higher than C16:0, C18:1 and total fatty acids compared with the control group. However, the ratio of n-6 to n-3 was three times lower than the control group. According to Alipanahi et al. (2019), adding oak fruit (containing hydrolyzable tannin) to the diet did not affect the pattern of ruminal fluid volatile fatty acids in lactating goats. Also, the diet containing oak fruit significantly increased the concentration of trans-vaccenic acid and unsaturated fatty acids and decreased the concentration of saturated fatty acids in goat milk fat.

Saponins, with protozoan release properties, change the rumen ecosystem and thus affect ruminal biohydrogenation. *Butyrivibrio fibrisolvens* (one of the bacteria involved in the biohydrogenation of unsaturated fatty acids) is able to break down saponins, and one of the reasons that saponins do not impact on unsaturated fatty acids may be the breakdown of saponins by ruminal microbes (Patra and Saxena, 2009).

Conclusions

Feeding tannins or saponins at the level used in the current study did not affect the performance criteria, ruminal pH and ammonia nitrogen concentration, and most blood constituents in Baluchi lambs fed the diet for 60 days. However, both saponin and tannin resulted in an increase in the number of white blood cells. There were significant differences in the concentration of individual fatty acids; however, the overall concentrations of the saturated and unsaturated fatty acids, and their ratios, were not influenced by tannin or

saponin treatment. The concentration of polyunsaturated fatty acids (PUFA) was highest in the lambs feeding on the tannic acid diet. Considering the limitation of this study, further research with different doses of tannins and saponins are needed before any recommendation can be made concerning the use of these compounds in fattening lambs.

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