

Research Article**Changes in lipid profile and some blood biochemical parameters in Karadi lambs receiving different levels of pomegranate peels**Karwan M Hama Khan¹, Hozan J Hamasalim¹, Sarwar M Sadq¹ and Dereen O.M. Ramzi²¹Department of Animal Production, Faculty of Agricultural Sciences, University of Sulaimani, Sulaymaniyah, Iraq;²College of Veterinary Medicine, University of Sulaimani, Sulaymaniyah, Iraq

<p>Article history Received: 9 Apr, 2015 Revised: 9 May, 2015 Accepted: 10 May, 2015</p>	<p>Abstract This study aimed to evaluate the effects of supplementing a basal diet with different levels of dietary pomegranate peels on some blood profile in Karadi lambs. Sixteen individual Karadi male lambs were used having age ranged between 4.5-5 months and average weight 23.29±0.42 kg. These lambs were divided into 4 groups. Basal diet was mixed with different levels of pomegranate peels powder at the rate of 0, 1, 2, 4% for 63 day. Diet was introduced <i>ad libitum</i> at 3% of live body weights twice a daily. At the end of the feeding period, biochemical analysis lipid profile (Total cholesterol, High-density lipoprotein (HDL), low-density lipoprotein (LDL), very low-density lipoprotein (VLDL), triglyceride and LDL/HDL ratio), total protein, serum albumin, serum globulin, blood urea nitrogen (BUN), creatinine and minerals (sodium, potassium, calcium, chloride, magnesium, Phosphorus, Iron and copper) were measured. The results indicated that supplementation of pomegranate peel in diet at the level of 1, 2 and 4% had a significant effect on LDL, ratio of LDL and HDL, albumin, creatinine, chloride, iron and copper in lambs. Keywords: Lipid profile, biochemical parameters, biochemical parameters, pomegranate peel, Karadi lamb</p>
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Introduction

Pomegranate is an important source of beneficial bioactive compounds and has been used for folk medicine for many centuries. Pomegranate fruit has been proved as a healthy fruit due to its biological actions. Pomegranate peel is nutritive rich by product which are abundant due to no more use. Pomegranate peels are being discarded after juice production and ready to eat arils. Pomegranate fruit is consisted of three parts: the seeds (about 3% of the weight of the fruit); the juice (about 30% of the fruit weight); and the

peels which include the husk and interior network membranes (Prakash and Prakash, 2011).

Hypercholesterolemia has been well known as a proven risk factor for cardiovascular disease (Wang et al., 2011). Hypercholesterolemia is generally, associated with an increase in plasma concentrations of low density lipoprotein (LDL-c) (bad cholesterol) and very low density lipoprotein (VLDL-c) and/or a decrease in high density lipoprotein cholesterol (HDL-c) (good cholesterol). Modification of oxidation of LDL-c is thought to play a key role during early atherogenesis i.e. formation of atheroma inside the

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walls of blood vessels that finally leads to arteriosclerosis (Kumar et al., 2008). Because of an increased resistance of LDL-c to oxidation after treatment with various synthetic pharmaceutical drugs (Breugnot et al., 1992; Pentikainen et al., 1995), there is a great need to identify natural food products that can offer antioxidant protection against LDL-c oxidation.

Pomegranate peel is a good source of antioxidant and thus may serve in the prevention of cattle diseases and improvement of beef products making it an attractive component in cattle feed. Shabtay et al (2008) demonstrated that dietary supplementation of pomegranate peels promotes an increase in feed intake, with a tendency to increase weight gain in bull calves. In contrast, Oliveira et al (2010) found that feeding a pomegranate extract to young calves for the first 70 d of life suppressed intake of grain and whole tract digestibility of fat and crude protein, likely because of its high tannin content. Recent studies also have shown that boosting antioxidant levels in the diet of cattle may help to improve their health. The peel packs some of the weight boosting and health enhancing effects of antibiotics and hormones without the detrimental effects and it may yield meat with higher level of beneficial antioxidants (Shabtay et al., 2008). Antioxidants also play an important role in food quality preservation due to their ability to prevent oxidative deterioration of lipids (Erukainure et al., 2012). The present study is undertaken to know the effect of using different ratios of pomegranate peels on lipid profile and some blood biochemical in Karadi male lambs.

Materials and Methods

Experiment animals

The experiment was conducted at farm of Animal Production Department, Faculty of Agricultural Sciences, University of Sulaimani and used 16 individual Karadi male lambs. Lambs were raised under similar environmental and management conditions. Lambs weighing 23.29 ± 0.42 kg and 4.5-5 months old were used. The lambs were purchased from a known local contractor and they were individually housed in pens (1 x 1.5 m) at the animal production farm and that allowed access to diets supplied in metal buckets, water was available at all time and the diets were gradually introduced to the lambs over a period of two weeks before the start of the experiment and all lambs were weighed weekly at 9.00 a.m. by hanging balance (100 gm sensitivity). Lambs were kept in clean and hygienic environment.

Preparation of pomegranate peels powder

Mature pomegranate fruits were washed and cut manually to separate the seeds and peel. The rind (peels) thus obtained, cut into small pieces using a

sharp knife and dried in an air circulatory tray drier at $60 \pm 5^\circ\text{C}$ for 6 h till its moisture content reached 12-14%. Dried pieces were cooled, powdered in a laboratory disc mill to pass through 20 mesh sieve, packed in high density polyethylene bags and stored at ambient temperature ($25 \pm 5^\circ\text{C}$) until use (Singh & Sethi, 2003; Devatkal & Naveena, 2010). The gross chemical components namely moisture, crude protein, crude fat, ash, crude fibres and carbohydrates content of pomegranate fruits peel powder are shown in Table 1.

Diets and chemical analyses

In this experiment, concentrate diets were used which contained: barely, yellow corn, soy bean meal, vitamins and minerals mixture. Barley straw was used as a source of roughage. Samples of feedstuff, offered feed and refusals were dried at 50°C until constant weight before chemical analyses. Samples then ground through a 1 mm screen for chemical analysis. Dry matter (DM), organic matter (OM), ether extract (EE), crude fibre (CF), nitrogen free extract (NFE), neutral detergent fibre (NDF) and acid detergent fibre (ADF) were determined according to FAO (2011).

The effect of four levels of pomegranate peels powder 0, 1, 2, 4% on the concentration and diurnal patterns of lipid profile (total cholesterol, HDL, LDL, VLDL, triglyceride and LDL/HDL ratio), total protein (TP), serum albumin, serum globulin, blood urea nitrogen (BUN) and creatinine was measured. In addition, sodium (Na), potassium (K), calcium (Ca), chloride (Cl), magnesium (Mg), phosphorus (P), iron (Fe) and copper (Cu) concentration at the end of experiment were also determined. Lambs in T1, T2, T3 and T4 were fed on a concentration diet with pomegranate peels powder at the rate of 0, 1, 2 and 4g/100 gm DM respectively. Formulation and chemical composition of the concentrate diets is shown in Table 2.

Sample collection and analysis

The blood samples were withdrawn from jugular vein from each lamb by using a disposable syringe into 10 ml plain tubes at the end of experiment (60 days). The test tubes were left to clot and then centrifuged at 3000 rpm for 20 min for serum separation and stored at -20°C until assayed for total HDL, LDL, VLDL, triglyceride, TP, serum albumin, serum globulin, BUN, creatinine, total bilirubin, Na, P, Ca, Cl, Mg, K, Fe and Cu using reagent kits (PZ CORMAY S.A., Poland) with Auto Biochemistry Analyzer (Model accent 200, Poland).

Statistical analysis

All data were subject to one-way analysis of variance (ANOVA) using XL Stat program for Windows. Differences between the means were tested by Fisher's Least Significant Difference (LSD) test.

The level of significance was chosen at $P < 0.05$ and the results were presented as mean (Steel et al., 1996).

Results

Lipid profile

The mean values (\pm SE) of lipid profile of control and experimental animals are shown in Table 3. The result of this study showed that different levels of pomegranate peel (1, 2, and 4%) had no significant effect on total cholesterol, HDL, VLDL and triglyceride concentration compared to control group. LDL concentration in lambs fed 4% pomegranate peel decreased significantly. Significantly lower LDL/HDL ratio was found in 4% pomegranate peel powder compared with control diet.

Biochemical parameters

Table 4 presents the mean values of some serum biochemical parameters in Karadi lambs. The result indicated that albumin and creatinin level increased significantly in group fed 4% pomegranate peels. No significant difference was found in concentration of total protein, globulin and BUN between control and treated groups.

Serum minerals

The levels of some serum mineral concentrations in blood of Karadi lambs are showed in Table 4. The result indicated that chloride concentration increased significantly in treated groups compared to control. Iron concentration decreased significantly in 2% pomegranate compared to control and treated groups. Copper concentration decreased significantly in 1%

pomegranate peel. No significant difference was found in other minerals in control and treated groups.

Table 1: Chemical composition of pomegranate peels (pp)

Item (%)	
Organic matter (OM)	96.2
Crude protein (CP)	5.1
Ether extract (EE)	4.9
Total Ash	3.7
Crude fiber (CF)	11.22
Nitrogen free extract (NFE)	80.50
Metabolizable energy ME (MJ/Kg)*	27.92

*ME was calculated according to Mirzaei-Aghsaghali (2011)

Table 2: Formulation and chemical composition of experimental diets

Ingredients (%)	T1	T2	T3	T4
	control	1%PP	2%PP	4%PP
Barley grain	40	40	40	40
Wheat barn	28	27	26	24
Yellow corn	20	20	20	20
Soybean meal	10	10	10	10
Pomegranate Peels	0	1	2	4
Salt	1	1	1	1
Minerals and vitamins	05	05	05	05
Dicalcium phosphate	05	05	05	05
Chemical composition%				
Organic matter (OM)	93.7	93.77	93.81	93.87
Crude protein (CP)	15.7	15.6	15.5	15.3
Ether extract (EE)	3.12	3.12	3.13	3.14
Total Ash	63	6.23	6.19	6.13
Crude fiber (CF)	7.8	7.7	7.6	7.5
Nitrogen free extract (NFE)	67.08	67.35	67.58	67.93
Metabolizable energy ME (MJ/Kg)*	12.63	12.65	12.67	12.69

*ME (MJ/ kg DM) = 0012 CP +0031 EE+0005 CF +0014 NFE (MAFF, 1977)

Table 3: Effect of dietary pomegranate peels on serum lipid profile in Karadi lambs (Mean \pm SE)

Parameters (mmol/l)	T1 (Control)	T2 (PP 1%)	T3 (PP 2%)	T4 (PP 4%)
T Cholesterol	1.64 \pm 0.05 ^a	1.55 \pm 0.33 ^a	1.41 \pm 0.02 ^a	1.61 \pm 0.16 ^a
HDL	0.65 \pm 0.07 ^a	0.58 \pm 0.02 ^a	0.69 \pm 0.03 ^a	0.92 \pm 0.05 ^a
LDL	1.10 \pm 0.10 ^a	1.31 \pm 0.54 ^a	0.61 \pm 0.02 ^a	0.24 \pm 0.00 ^b
VLDL	0.15 \pm 0.14 ^a	0.16 \pm 0.02 ^a	0.14 \pm 0.24 ^a	0.15 \pm 0.00 ^a
Triglyceride	0.23 \pm 0.04 ^a	0.36 \pm 0.13 ^a	0.52 \pm 0.17 ^a	0.28 \pm 0.02 ^a
LDL/HDL ratio	1.69 \pm 0.01 ^a	2.22 \pm 0.86 ^a	1.14 \pm 0.14 ^a	0.25 \pm 0.01 ^b

Control = basal diet, PP 1% = basal diet+1% pomegranate peels, PP 2% = basal diet+2% pomegranate peels, PP 4% = basal diet+4% pomegranate peels. Mean values with different superscripts differ significantly ($P < 0.05$)

Table 4: Effect of dietary pomegranate peels on some blood biochemical parameters in Karadi lambs (Mean \pm SE)

Parameters (mg/dl)	T1 (Control)	T2 (PP 1%)	T3 (PP 2%)	T4 (PP 4%)
T Protein	6.44 \pm 1.47 ^a	5.74 \pm 0.72 ^a	6.53 \pm 0.63 ^a	6.65 \pm 0.28 ^a
Albumin	3.14 \pm 0.69 ^b	3.15 \pm 0.00 ^b	3.22 \pm 0.05 ^{ab}	3.27 \pm 0.02 ^a
Globulin	3.30 \pm 2.16 ^a	2.59 \pm 0.72 ^a	3.31 \pm 0.69 ^a	3.37 \pm 0.26 ^a
B UN	5.40 \pm 0.23 ^a	5.20 \pm 0.57 ^a	5.11 \pm 0.16 ^a	4.7 \pm 0.02 ^a
Creatinine	0.61 \pm 0.01 ^b	0.73 \pm 3.55 ^{ab}	0.77 \pm 5.34 ^a	0.79 \pm 0.01 ^a

Control = basal diet, PP 1% = basal diet+1% pomegranate peels, PP 2% = basal diet+2% pomegranate peels, PP 4% = basal diet+4% pomegranate peels. Mean values with different superscripts differ significantly ($P < 0.05$).

Table 5: Effect of dietary pomegranate peels on some serum mineral parameters in Karadi lambs (Mean ± SE)

Parameters (mmol/l)	T1 (Control)	T2 (PP 1%)	T3 (PP 2%)	T4 (PP 4%)
Sodium	146.50±0.28 ^a	145.5±0.28 ^a	145.0±2.30 ^a	147.16±1.42 ^a
Potassium	4.60±0.05 ^a	4.45±0.14 ^a	4.55±0.37 ^a	4.90±0.17 ^a
Calcium	1.43±0.09 ^a	1.39±0.02 ^a	1.58±0.20 ^a	1.27±0.02 ^a
Chloride	99.0±0.57 ^b	101.8±1.48 ^a	104.0±0.57 ^a	102.50±0.28 ^a
Magnesium	0.02±0.00 ^a	0.26±0.19 ^a	0.07±0.04 ^a	0.04±0.02 ^a
Phosphorus	1.87±0.10 ^a	2.03±0.09 ^a	1.97±0.34 ^a	2.34±0.17 ^a
Iron	36.15±2.62 ^{ab}	42.05±2.51 ^a	22.15±1.53 ^c	31.70±1.44 ^b
Copper	21.25±0.02 ^{ab}	19.30±1.44 ^b	22.0±1.32 ^{ab}	23.65±1.06 ^a

Control = basal diet, PP 1% = basal diet+1% pomegranate peels, PP 2% = basal diet+2% pomegranate peels, PP 4% = basal diet+4% pomegranate peels. Mean values with different superscripts differ significantly (P<0.05)

Discussion

In our study, the results clearly show that the pomegranate peel has no effect on total cholesterol concentration in Karadi lambs. The result is in agreement with Hussein and Shujaa (2013) who reported that different levels (2, 4 and 6%) of pomegranate peel had no any effect on cholesterol concentration in Awassi lambs. Also total cholesterol concentration in this study was within the reference range as reported by Hama Khan et al. (2013). However, these results are in disagreement with the results of Hossin (2009) who reported that different levels of pomegranate peel significantly increased total cholesterol in rats. The obtained results are inconsistent with data of Osman et al. (2012) who found that oral administration of pomegranate peel led to significant increase in cholesterol level in rats. This disparity could stem from the genotype, age and physiological status of the experimental animals. In the current study, different levels of pomegranate peel powder did not alter serum HDL density. The mean value for this parameter was in the normal range as reported for Karadi sheep (Hama Khan et al., 2013). Like our results, Hossin (2009) illustrated no significant difference in HDL in rats fed on diet with three different level of pomegranate peel (5, 10 and 15%). This contradict with study of Osman et al. (2012) who showed that pomegranate peel powder decreased serum HDL in rats fed on diet supplemented with pomegranate. Results of the present study suggested that supplementing inedible part of pomegranate can decrease concentration of LDL. Mean of these parameters are within the reference range as found for Karadi sheep (Hama Khan et al., 2013). Lambs fed on diets supplemented with high level of pomegranate peel reduced LDL concentration as compared with control diet. In contrast, Osman et al. (2012) revealed an increase in LDL in serum of rats fed on diet supplemented with pomegranate peel compared to rats kept as control group. The results of this experiment were consistent with previous reports that diets supplemented with different levels of pomegranate

had no effect on VLDL concentration compared with control group (Hossin, 2009). Similarly, Hussein and Shujaa (2013) did not find any difference in triglyceride concentration in Awassi lambs due to pomegranate peel supplementation. Moreover, Hossin (2009) who reported that dietary supplementation with pomegranate peel has no significant effect on triglyceride concentration in rats compared with control group. The mean value of triglyceride concentration in this study is within the reference range as reported for Karadi sheep in Sulaimani city (Hama Khan et al., 2013).

Limited data are available on effect of pomegranate peel on biochemical parameters in lamb. In the present study, pomegranate peel had detectable effects on serum albumin concentration in Karadi lambs. In the present work, it was clear that lambs fed on diet supplemented with different levels of pomegranate peel had no significant effect on blood urea nitrogen. These results are contrary to the results of Hussein and Shujaa (2013) who reported different levels of pomegranate peel (2, 4 and 6%) significantly decreased blood urea nitrogen in Awassi lambs. The study has shown that there are statistically significant differences in some mineral concentration between treatments of Karadi lambs. To the best of our knowledge, no information is available on serum mineral concentration in response to pomegranate supplementation. These values may help as a base for future research on lambs.

Conclusion

In our study, administration of pomegranate peel in diet at the level of 1, 2 and 4% had a significant effect on LDL, ratio of LDL and HDL, albumin, creatinine, chloride, iron and copper in lambs.

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