



Impact of Feeding Tea Leaves Saponins on Some Productive and Serum Biochemical Parameters of Awassi Lambs

Rasheed H. Abdullah & Hanaa A. J. Al-Galbi*

Department of Animal Production, College of Agriculture, University of Basrah, Iraq

*Corresponding author: hanaa.jaba@gmail.com

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Abstract: The study was conducted in one of the private fields of Wasit Governorate for the period from 22/1/2019 to 11/5/2019. The study aimed to investigate the impact of feeding different levels of saponins on the productive performance of Awassi lambs. Sixteen Awassi lambs aged three-four months with an average weight of 23.68 kg were used. The lambs were randomly divided into four experimental treatments with four animals for each. The diet provided at 3% of the body weight. The feed was provided with two meals (8 am and 4 pm). The 1st treatment was control (without addition), while in the 2nd, 3rd, and 4th treatments, saponins was added at level of 60, 120 and 180 mg.kg⁻¹ DM feed respectively. Adding saponins at 180 mg.kg⁻¹ DM increased the final body weight at the second and third months of the study with 34.35 and 40.36 kg, respectively. Total gain was increased in this treatment from 15.03 kg in control to 17.47 kg. Feed conversion ratio of the same treatment was 5.52 as compared with 6.24 kg.kg⁻¹ feed for the control treatment. Glucose and protein concentrations in the serum also showed an improvement due to the addition of saponins. Values were, 57.24 mg.100 ml⁻¹ and 4.83 g.100 ml⁻¹ respectively. Those of control group were 51.86 mg.100 m⁻¹ and 4.62 g.100 ml⁻¹ respectively. Cholesterol concentration in the serum was not affected by addition of different levels of saponins. It can be concluded that addition of saponins at 180 mg. kg⁻¹ feed improved body weight, gain, feed conversion ratio and some serum biochemical parameters.

Keywords: Awassi lambs, Serum, Saponins, Gain, Conversion.

Introduction

General concerns about chemical compounds that are used as feed additives have increased, and in this context safer alternatives have been found including the use of secondary compounds produced by plants such as saponins, essential oils, tannins, etc. (Khalifa *et al.*, 2014). There is a growing interest by researchers in the field of nutrition about the

use of plant extracts as natural additives in their efforts to modify rumen fermentation and make it more efficient in digesting fiber and improving the efficiency of feed use or reducing protein degradation and thus increasing its flow to the duodenum, improving the quality of meat, improving nutrient digestibility while reducing methane and

ammonia production as well as emissions (Sliwinski 2004; Szumacher-Strabel & Cieślak, 2010). Several measures have been considered to manipulate the production of gases, as plant extracts have been found to have more integrated benefits or are considered to be an effective way to reduce methane and ammonia emission compared to chemical additives such as antibiotics. Plant extracts can be combined as a growth enhancer or anti-inflammatory, fungus, infection and poisoning (Sun *et al.*, 2017).

Among the nutritional solutions developed are the use of saponins, which are usually high in tropical plants, promising natural materials to reduce ammonia and methane emissions and to modify rumen fermentation (Budan *et al.*, 2013).

Liu *et al.* (2018) found significant difference in final body weight when feeding lambs diets with saponins at different levels (2000 and 4000) mg.kg⁻¹ dry matter (42.5 and 42.4 kg, respectively), compared with the control treatment which recorded 39.6 kg. Liu *et al.* (2019) indicated when adding two levels of saponins (0 and 2.0 g. head⁻¹ per day to sheep diets, there was a significant increase in the final weight compared to the control treatment. However, adding saponins at different levels did not significantly affect the amount of feed consumed (Liu *et al.*, 2018; 2019). Aazami *et al.* (2013) indicated that adding saponins at different levels (0, 100 and 200 mg. kg⁻¹ dry matter) to sheep diets, there were no significant differences in the concentration of both glucose and triglycerides in all levels of addition while a significant decrease was found in cholesterol concentration in the third treatment compared with the control treatment.

The current study aimed to investigate the impact of feeding Awassi lamb different levels of tea seed saponin on body weight, gain and some serum biochemical parameters.

Materials & Methods

Extraction of saponins from tea leaves

Saponin from tea leaves was extracted following the method developed by Hussien & Awad (2014). In brief, tea leaves were collected, gently cleaned, dried, then grinded. Dissolve 30 g of crushed powder with 450 ml of 99.8% methanol for a period of 7 days under room temperature. After filtering, the extracted liquid evaporated under low pressure in a rotary evaporator until thick residual is formed. The residual was then washed with petroleum ether to remove chlorophyll and fatty substances, and the process continued until the residual became colorless. Dissolving the residual is repeated in 100 ml of 99.8% methanol and added to the resulting solution, diethyl ether. A yellowish residual will be formed from saponins and the process of adding diethyl ether will continue until the sedimentation process was stopped. The resulted compound was dried by continuous stirring at the laboratory temperature to produce the raw extract which was estimated to be 4.3 g of saponins.

Productive traits

Body weight (kg) was recorded by normal balance for lambs. Growth rate gain (kg) was estimated as the difference between initial and final body weights. Total feed consumption (kg) was the difference between offered feed and the residual. Feed conversion ratio was estimated by dividing feed consumed on total gain (kg.kg⁻¹).

Serum biochemical parameters

Serum glucose, total protein, cholesterol and triglycerides were determined using kit produced by French company (BIOLABO SA) as:

$$\text{Glucose (mg/100 cm}^3\text{)} = \left(\frac{\text{sample reading}}{\text{standard solution}} \right) * 5.32$$

$$\text{Total protein (g/100 cm}^3\text{)} = \left(\frac{\text{sample reading}}{\text{standard solution}} \right) * 6$$

$$\text{Cholesterol (mg/100 cm}^3\text{)} = \left(\frac{\text{sample reading}}{\text{standard solution}} \right) * 200$$

$$\text{Triglycerides (mg/100 cm}^3\text{)} = \left(\frac{\text{sample reading}}{\text{standard solution}} \right) * 200$$

Feeding groups

Animals were distributed into four feeding treatments. The first treatment was a control (no addition), in which diet included 50% barley, 20% wheat bran, 20% corn, 7.5% soya bean meal and 2.5% vitamin and minerals. The second treatment was a control ration plus 60 mg.kg⁻¹ feed of tea saponin. The third treatment was a control ration plus 120 mg.kg⁻¹ feed of tea saponin. The fourth treatment was a control ration plus 180 mg.kg⁻¹ feed of tea saponin. Wheat straw was offered ad libitum for all treatments.

Statistical analysis

Data were analyzed according to a completely randomized design. Treatments effects on studied traits were analyzed using one-way ANOVA design (SPSS, 2016, version 24). The significant differences among means were compared by least significant test within the same statistical packages. Significant level considered as 0.05. Each treatment had three replicates.

Results & Discussion

Final weight and total gain

Table (1) shows significant differences (P<0.05) among the means of both final weights and total gain of lambs in the experimental treatments. Statistical analysis showed no significant differences in the final weight and gain for all treatments in the first month. The significant differences were existed (P<0.05) in the second and third months of the experiment, where the fourth treatment (saponins at a level of 180 mg) in the second month recorded higher significant increase (P<0.05) in the final weight (34.35 kg) as compared with the first and second treatments (31.70 and 31.90 kg, respectively). Results also showed a significant superiority (P<0.05) for the second, third and fourth treatments for which saponins were added at level of 60, 120 and 180 mg.kg⁻¹ feed, respectively, in the final weight at the third month (38.50, 39.83 and 40.36 kg respectively) as compared with the control (37.93 kg). The fourth treatment

recorded higher ($P < 0.05$) significant increase (17.74 kg) in total gain as compared with the control and second treatments (15.03 and 15.66 kg, respectively). However, adding saponin at 120 mg.kg⁻¹ showed similar gain as that of 180 mg.kg⁻¹ (16.69 kg). the improvement in both body weight and total gain may be due to the effective role of saponins in improving the rumen condition by providing a suitable pH for the growth of microorganisms that improve the digestion and absorption of nutrients. Saponin also may serves to reduce ammonia and thus increases the flow of protein to the intestine, and as a result improves body weight and weight gain. These results are consistent with

Liu *et al.* (2018), they used saponins at different levels (0, 500, 1000, 2000 and 4000) mg. kg⁻¹ dry matter, and found a significant increase ($P < 0.05$) in the final weight of the fourth and the fifth treatments as compared with other treatments. Liu *et al.* (2019) also observed a significant increase ($P < 0.05$) in the final weight of sheep due to feeding diets containing 2 g. head⁻¹ per day of saponins as compared with the control treatment. However, Mao *et al.* (2010) and Nasri *et al.* (2011) did not observe significant differences in the final weight and the gain due addition of saponins to lambs' diets.

Table (1): Effect of adding different levels of saponins on body weight and the total gain (kg. head⁻¹) of Awassi lambs at different periods (Mean± SD).

Treatments (Saponin level)	Body weight (kg)				Total gain (kg)
	Initial	1 st month	2 nd month	3 rd month	
0 mg.kg ⁻¹ feed (Control)	22.90 ± 1.21	26.92 ± 0.13	31.70 ^b ± 0.30	37.93 ^b ± 0.90	15.03 ^b ± 0.50
60 mg.kg ⁻¹ feed	22.83 ± 0.28	27.13 ± 0.41	31.90 ^b ± 0.60	38.50 ^a ± 0.78	15.66 ^b ± 0.49
120 mg.kg ⁻¹ feed	23.14 ± 1.39	27.40 ± 1.24	33.30 ^{ab} ± 1.68	39.83 ^a ± 1.89	16.69 ^{ab} ± 0.81
180 mg.kg ⁻¹ feed	22.50 ± 1.01	27.33 ± 0.62	34.35 ^a ± 0.58	40.36 ^a ± 1.21	17.74 ^a ± 0.65

- Vertically different letters mean significant differences at $P < 0.05$ level.

Feed consumption and feed conversion ratio

Table (2) shows the amount of feed consumed and the feed conversion ratio. Statistical analysis was not performed due to group feeding. Highest amount of feed was consumed by lambs in the fourth treatment throughout the trial period (97.98 kg) followed by the third (97.13 kg). The least

amount of feed was consumed by lambs the first (control) and second treatments throughout the trial period (93.82 and 94.52 kg, respectively).

Regarding feed conversion, the table showed that the best value was recorded by lambs in the fourth treatment (5.52) followed by those in the third treatment (5.82 kg feed.kg⁻¹ weight gain). In both

treatments feed conversion ratio was better than that recorded by lambs in the first and the second treatments (6.24 and 6.03 kg feed.kg⁻¹ weight gain) respectively. This may be due to the fact that addition of saponin improved the rumen condition, by stimulation the efficiency of microbial growth and the inhibition of protein degradation in the rumen, which in turn has been reflected in the efficiency of utilization

of the metabolizable protein reaching the intestine. The results were not in consistent with those reported by Mao *et al.* (2010) and Nasri *et al.* (2011), in which no differences in the amount of feed consumed and the feed conversion ratio due to addition of saponins to lambs' diets. Liu *et al.* (2018 & 2019) found no significant effect of saponin on feed consumption and feed conversion ratio by sheep.

Table (2): Effect of adding different levels of saponins on feed consumed and feed conversion ratio.

Treatments (saponin level)	Feed consumption (kg)	Feed conversion ratio (kg feed.kg ⁻¹ weight gain)
0 mg.kg ⁻¹ feed (Control)	93.82	6.24
60 mg.kg ⁻¹ feed	94.53	6.03
120 mg.kg ⁻¹ feed	97.14	5.82
180 mg.kg ⁻¹ feed	97.99	5.52

There are no significant differences among different treatment means.

Serum biochemical parameters

Glucose

Table (3) shows that there were no significant differences (P<0.05) in the mean blood glucose concentration among experimental treatments due to addition of saponins at the following levels (0, 60, 120 and 180) mg.kg⁻¹ dry matter at the first month. Values were 50.41, 51.27, 52.38 and 51.14 mg. (100 cm³)⁻¹, respectively.

However, the table shows the presence of significant differences (P<0.05) in the mean serum concentration of glucose at the second month. The fourth treatment was

significantly superior (P<0.05) with 57.24 mg.100⁻¹ cm³ as compared with the 1st, 2nd and 3rd treatments, values were 51.86, 53.13, and 53.60 mg.100⁻¹ cm³, respectively. Table (3) also shows the presence of significant differences (P<0.05) in the serum blood glucose concentration for the third, and the fourth treatments (59.00 and 60.61 mg.100⁻¹ cm³, respectively) as compared with the first and second treatments (55.01 and 55.09 mg.100⁻¹ cm³, respectively). These results were not in agreement with those obtained by Aazami *et al.* (2013) due to addition of saponins at different levels (0, 100 & 200) mg. kg⁻¹ dry matter to sheep diets, where,

they noticed no significant differences ($P < 0.05$) in the concentration of glucose in all levels of addition.

Total protein

Results of table (3) shows the presence of significant differences ($P < 0.05$) in the concentration of protein among the different experimental treatments. The fourth treatment was significantly superior

($P < 0.05$) at the first month ($4.83 \text{ g.100 cm}^3^{-1}$) as compared with the first & second treatments (4.65 and $4.62 \text{ g.100 cm}^3^{-1}$), respectively). Whereas, the third treatment did not differ significantly ($4.73 \text{ gm.100 cm}^3^{-1}$) as compared with other experimental treatments at the first month. Table (3) also shows that there were no significant differences ($P < 0.05$) in the serum protein concentration among different experimental treatments at the second and third months.

Table (3): Effect of adding different levels of saponins on serum glucose and total protein concentrations at different periods (Mean \pm SD).

Treatments (Saponin level)	Glucose concentration mg. (100 cm ³) ⁻¹			Total protein concentration g.100 cm ³) ⁻¹		
	1 st month	2 nd month	3 rd month	1 st month	2 nd month	3 rd month
0 mg.kg ⁻¹ feed (Control)	50.41 ± 2.04	51.86 ^b ± 1.45	55.01 ^b ± 1.37	4.62 ^b ± 0.07	4.95 ± 0.12	5.01 ± 0.32
60 mg.kg ⁻¹ feed	51.27 ± 1.86	53.13 ^b ± 3.12	55.09 ^b ± 1.98	4.65 ^b ± 0.08	5.02 ± 0.24	5.05 ± 0.23
120 mg.kg ⁻¹ feed	52.38 ± 3.05	53.60 ^b ± 3.12	59.00 ^b ± 0.90	4.73 ^{ab} ± 0.01	5.25 ± 0.34	5.19 ± 0.59
180 mg.kg ⁻¹ feed	51.14 ± 2.39	57.24 ^a ± 1.16	60.61 ^a ± 1.95	4.83 ^a ± 0.09	5.45 ± 0.50	5.22 ± 0.51

- Vertically different letters mean significant differences at $P < 0.05$ level.

Cholesterol and triglycerides

Results presented in table (4) indicated that there were no significant differences in the serum concentration of both cholesterol and triglycerides among different experimental treatments as affected by addition of saponins at different levels (0, 60, 120, and 180 mg.kg⁻¹ dry matter). At the first month, serum concentrations of cholesterol were 53.60, 51.83, 50.30, and 50.40 mg. (100 cm³)⁻¹, respectively. While the values of

triglycerides were 63.23, 62.04, 62.48, and 63.79 mg.100⁻¹ cm³ respectively. Table (4) also shows that there were no significant differences in the serum concentrations of both cholesterol and triglycerides at the second and third months among different experimental treatments due to addition of saponins. These results are similar to those obtained by Aazami *et al.* (2013) when they added saponins at different levels (0, 100

and 200) mg.kg⁻¹ dry matter to sheep diets and no significant differences were found in

the serum concentration of triglycerides.

Table (4): Effect of adding different levels of saponins on serum cholesterol and triglyceride concentrations in lamb serum at different periods (Mean ±SD).

Treatments (Saponin level)	Cholesterol (mg.100 cm ³⁻¹)			Triglyceride (mg.100 cm ³⁻¹)		
	1 st month	2 nd month	3 rd month	1 st month	2 nd month	3 rd month
0 mg.kg ⁻¹ feed (Control)	53.60 ±6.78	56.29 ±5.50	58.74 ±3.59	63.23 ±3.37	66.13 ±2.44	67.53 ±1.54
60 mg.kg ⁻¹ feed	51.83 ±4.14	55.06 ±2.70	57.10 ±3.40	62.04 ±2.60	65.67 ±2.26	66.67 ±3.68
120 mg.kg ⁻¹ feed	50.30 ±4.46	54.78 ±4.20	55.16 ±3.68	62.48 ±6.17	65.38 ±5.00	66.26 ±4.80
180 mg.kg ⁻¹ feed	50.40 ±1.45	53.76 ±6.52	55.36 ±5.09	63.79 ±3.51	64.56 ±3.59	65.50 ±2.65

- There are no significant differences among different treatment means.

Conclusion

It can be concluded that adding saponins at 180 mg.kg⁻¹ dry matter increases the body weight at the second and third months of the study (34.35 and 40.36 kg, respectively) and gain (17.47 vs 15.03 kg for control). Feed conversion ratio of the 180 mg.kg⁻¹ of saponin treatment was 5.52 vs. 6.24 kg feed.kg⁻¹ for the control treatment. Serum glucose and protein concentrations also showed an improvement with the addition of saponin specially at level of 180 mg.kg⁻¹. The values were 57.24 and 4.83 g (100 cm³)⁻¹ respectively. Those of control group were 51.86 and 4.62 g.100 cm³⁻¹ respectively.

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Conflicts of interest

The authors-declare-that they-have-no-conflict of interests.

Ethical approval

All applicable institutional, national and international guidelines for the care and use of animals were followed.

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تأثير صابونين ورق الشاي على بعض المعايير الانتاجية والبيوكيميائية في مصل الدم في الحملان العواسية

رشيد حميد عبد الله و هناء علي جبار الغالبي•

قسم الانتاج الحيواني، كلية الزراعة، جامعة البصرة، العراق

المستخلص: أجريت الدراسة الحالية في أحد الحقول الخاصة بمحافظة واسط للفترة من 2019/1/22 إلى 2019/5/11. هدفت الدراسة إلى معرفة أثر تغذية مستويات مختلفة من الصابونين على الأداء الإنتاجي للحملان العواسي. تم استخدام ستة عشر حمل عواسي تتراوح أعمارهم بين 3-4 أشهر بمتوسط وزن 23.68 كجم. تم تقسيم الحملان بشكل عشوائي إلى أربعة معاملات تجريبية، أربعة حيوانات لكل منها. قدمت العليقة بنسبة 3 % من وزن الجسم. تم تقديم العلف خلال وجبتين (الأولى عند 8 صباحاً والثانية عند 4 مساءً). كانت المعاملة الأولى هي السيطرة (بدون إضافة)، أما المعاملات الثانية والثالثة والرابعة، فتمت إضافة الصابونين عند مستوى 60 و 120 و 180 ملغم.كغم⁻¹ من المادة الجافة للعليقة (DM) على التوالي. أدت إضافة الصابونين عند 180 ملغم.كغم⁻¹ DM إلى زيادة وزن الجسم النهائي في الشهر الثاني والثالث من الدراسة (34.35 و 40.36 كجم على التوالي). تم تحسن الزيادة الوزنية الكلية في هذه المعاملة من 15.03 كغم إلى 17.47 كغم. كانت كفاءة التحويل الغذائي لنفس المعاملة 5.52 بالمقارنة مع 6.24 كجم.كغم⁻¹ لمعاملة السيطرة. كما أظهر تركيز الكلوكوز والبروتين في المصل تحسناً بسبب إضافة الصابونين وكانت قيمهما 57.24 ملغم. 100 مل⁻¹ و 4.83 غم. 100 مل⁻¹ على التوالي. بينما سجلت مجموعة السيطرة 51.86 ملغم. 100 مل⁻¹ و 4.62 غم. 100 مل⁻¹ على التوالي. لم يتأثر تركيز الكوليسترول في المصل بإضافة مستويات مختلفة من الصابونين. يمكن الاستنتاج، ان إضافة الصابونين بمقدار 180 ملغم.كغم⁻¹ يمكن ان يحسن من وزن الجسم والزيادة الوزنية الكلية وكفاءة التحويل الغذائي وبعض المعايير البيوكيميائية في الدم.

الكلمات المفتاحية: حملان العواسي، المصل، الصابونين، الزيادة الوزنية، كفاءة التحويل الغذائي.