Effect of Level of Concentrate Feeding and Addition of Monensin on Blood Parameters of Awassi Lambs

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Abstract

This study was conducted to investigate the effect of level of concentrate feeding and addition of monensin on blood parameters. Sixteen Awassi lambs were used at 4-6 months of age and mean initial weight of 21.27 kg. Concentrate diet was offered at two levels (2.5 and 3% of body weight, BW) with or without the addition of monensin at a rate of 30 mg/kg DM. Ground wheat straw was offered \textit{ad libitum}. Results showed that increasing level of concentrate significantly increased (P<0.01) blood concentration of total protein (BTP) from 6.94 to 8.84 g.100 ml\(^{-1}\), urea nitrogen (BUN), and triglyceride (TG) from 15.54 to 18.23 and 22.66 to 25.12 mg.100 ml\(^{-1}\) respectively. The addition of monensin significantly increased (P<0.05) TP concentration from 7.61 to 8.17 g.100 ml\(^{-1}\), whereas, BUN concentration was decreased (P<0.01) from 18.15 to 15.62 mg.100 ml\(^{-1}\). All blood parameters were also affected (P<0.05) by the interaction between the level of concentrate and the addition of monensin. In the study of diurnal changes, blood parameters showed an expected response to the time of withdrawing blood samples from lambs.

Keyword: Monensin, Concentrate, Blood parameters.

1. Introduction

Protein supplements, feed additives such as medicinal plants or probiotics and monensin are very important materials that can improve growth rate, feed efficiency utilization, and carcass characteristics of Awassi lambs [1]. Additives, such as monensin were reported to influence blood constituents through remodel of ruminal microbial populations, which was reflected by changes in fermentation end products [2], i.e. the increase of propionic acid was associated with increased plasma glucose concentration, and increased ruminal molar proportion of butyrate was associated with raised plasma BHBA concentration. According to Stradiotti Júniör, et. al., [3], ionophores act on ruminal microbes and inhibit gram-negative species, these bacterial species are the main responsible for amino acid deamination and produce unwanted gases, such as methane and ammonia. The inhibition of these bacteria increases the production of propionate and the levels of blood glucose [4], [5] reported that monensin has the potential to increase the supply of glucogenic precursors, thereby increasing the hepatic synthesis of glucose and consequently improving energy balance. This improvement was associated with changes in some blood parameters such as insulin and growth hormone [6], blood urea and uric acid [7], total protein and blood sugar [8]. Therefore, this study aimed to evaluate the effect of introducing monensin in concentrate diet offered to lambs at low and high levels on blood parameters.

2. Material and Methods

This study was carried out at the animal field/Animal Production Department- College of Agriculture- Al-Qasim Green University from 28/11/2018 to 16/3/2019. The study included feeding Awassi lambs two levels of concentrate diet 2.5 and 3% of BW with or without the addition of monensin. Thus there will be 4 experimental treatments. A concentrate diet was prepared by mixing several ingredients including wheat bran, barley, yellow corn, and soybean meal. In addition to that, salt and minerals- vitamin mix was offered as well. The level of each ingredient was estimated to ensure that the concentrate diet contains ~12.5% CP and 1.34 g of rumen degradable nitrogen (RDN)/each mega joule of metabolizable energy (g RDN.MJ\(^{-1}\) of ME). The estimated content of RDN was ~1.64 g.100 g DM\(^{-1}\) and content of ME was about 1.23 MJ.100 g DM\(^{-1}\), then, the estimated ratio of RDN/ME was 1.34. The above estimations were performed according to the effective ruminal degradability of protein fraction in the ingredients used in the concentrate diet as mentioned in table

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1. Monensin as 10% monensin sodium was added at level of 30 mg.kg DM⁻¹. The chemical composition of the concentrate diet, ingredients, and wheat straw is shown in table 1.

Sixteen Awassi male lambs were used in this study. Lambs were bought from the local market with an initial average weight (21.275 ± 2.56 kg) and age ranged (4-6 months). Lambs were randomly divided into four treatments, 4 lambs each and housed individually. Lambs were well adapted to individual cages and study conditions before the beginning of the study. Concentrate diets were offered gradually with two meals at 8 am and 4 pm.

In the last week of the study, blood samples were withdrawn from the Jugular vein from each lamb. Samples were transferred to sterile tubes containing anticoagulant before morning feeding (zero time), and after 3, and 6 hrs following determine BG, BTP, BUN, and BTG concentrations. Blood samples were centrifuged and separated serum was collected and stored at -20°C until analysis. Spectrophotometrically using SP-3000 UV-Visible Spectrophotometer and commercial ready-made solutions kit.

Data obtained were statistically analyzed according to factorial experiments (2 × 2) in a completely randomized design (CRD) to evaluate the effect of the main factors studied in the experiment. Statistical Analysis System. SAS [14] was used for that purpose.

3. Results and Discussion

3.1. Effect of level of concentrate feeding and the addition of monensin on blood parameters

The effect of concentrate levels and the addition of monensin on blood parameters are presented in table 2. Results revealed that increasing the level of concentrate from 2.5 to 5% of BW of the Awassi lambs’ diet had no significant effect on BG concentration (66.02 - 65.69 mg.100 ml⁻¹). Insignificant effect on BG concentrations (67.14 vs. 70.92 mg.100 ml⁻¹) was due to an increasing level of concentrate from 2 to 3% of BW in the lambs’ diet shown by Keady and Hanrahan [15].

A similar result was obtained by Vosooghi-poostindoz, et. al., [16] who observed that the BG concentration of lambs was not significantly affected (58.0 vs. 58.66 mg.100 ml⁻¹) by increasing concentrate level in the lamb diet. In another study, increasing the level of concentrate from 2.73 to 3.46 % of BW of the Awassi wethers diet had no significant effect on BG concentrations [17]. The insignificant effect of concentrate levels on blood glucose in a current study might be explained by the relatively small number of experimental lambs and insufficient statistical power of the experiment.

Results of a current study revealed that increasing level of concentrate from 2.5 to 3% of BW increased (P<0.01) BTP from 6.94 to 8.84 g.100 ml⁻¹, BUN from 15.54 to 18.23 mg.100 ml⁻¹, and BTG from 22.66 to 25.12 mg.100 ml⁻¹. Allam, et. al., [18] concluded that higher BTP concentration may have resulted from higher intake and digestion of protein.

In agreement with the result of the current study, Can et. al., [19] found that increasing the level of concentrate from 2.43 to 2.84% of BW in the lamb’s diet increased (P<0.05) BUN concentrations from 12.35 to 23.00 mg.100 ml⁻¹. Similarly, Vosooghi-poostindoz and his coworkers [16] showed an increase (P<0.05) in BUN concentrations from 12.3 to 16.3 mg.100 ml⁻¹ as a result of increasing level of concentrate from 1.89 to 2% of BW in the lambs.

Higher BUN concentration in the lambs fed high concentrate may be related to ruminal ammonia-N concentration. The findings of Azizi-Shotorkhoft, et. al., [17] have shown a positive correlation between BUN ruminal NH₃-N concentrations. The increased BUN concentrations can be explained largely by increased absorption of ruminal ammonia, resulting in greater quantities of ammonia being detoxified in the liver to form urea. Moreover, since urea is excreted in urine and milk, then urea-N in the blood will be associated with nitrogen use efficiency, its concentration is increased by increasing protein consumption and by increasing the decomposing protein in the rumen [20].

With regard to the effect of the concentrate level on BTG, Dosky [21] reported a similar result as that previously mentioned in the current study. Russell [22] attributed higher BTG concentration associated with higher concentrate level and to the higher content of soluble sugars, where there is a direct relationship between carbohydrates and fats often exist.

Regarding the effect of the addition of monensin at a rate of 30 mg.kg DM⁻¹ on blood parameters, results revealed that there was no significant effect on BG, 64.09 vs. 67.62 mg.100 ml⁻¹. A similar result was shown in another study in which Reis, et. al., [23] found that concentrations of BG were insignificantly influenced as a result of the addition of monensin at a rate of 33 mg.kg DM⁻¹ diets.

In concern with the insignificant effect of monensin on blood glucose observed in a current study, Duffield, et. al., [24] reported that this case is most likely a function of the response being small and generally requiring a larger sample size to effectively assess the glucose response. Though it was not significant, the increase in BG concentration from 64.09 to 67.62 mg.100 ml⁻¹ in a current study may support that attribution.

Results of a current study also revealed that BTP concentration was significantly (P<0.05) increased from 7.61 to 8.17 g.100 ml⁻¹ due to the addition of monensin. The positive effect of monensin supplementation on BTP was attributed to increased protein digestion post rumen through the effect of proteolytic enzymes and changes in the amino acid profile of feed portions due to increased microbial protein synthesis [25].
Regarding BUN, the results of a current study showed that the addition of monensin to Awassi lambs diet decreased (P<0.01) BUN concentration from 18.15 to 15.62 mg.100 ml⁻¹. Similar results were observed by Khoshadi, et. al. [26] who reported that the monensin additive at the levels of 0, 20, and 40 mg.kg DM⁻¹ to Zeal lambs diets, resulted in a significant decrease (P<0.05) in BUN concentration from 14.49 to 10.33 and 11.99 mg.100 ml⁻¹. A significant decrease (P<0.05) in BUN concentration from 54.7 to 44.2 mg.100 ml⁻¹ was also recorded by Asadi, et. al. [27] due to the inclusion of monensin into Farahani lamb's diets at a rate of 24 mg.kg DM⁻¹ respectively. Worth mentioning, the plasma urea-N concentration is related to the level of ammonia absorption from the rumen and/or the deamination of amino acids not deposited in the tissue [28]. Ding, et. al., [29] reported that the lower plasma urea-N concentration may be attributed to the role of monensin to promote the utilization and deposition of nitrogen in tissues.

Results in the current study revealed that the addition of monensin slightly increased BTG from 23.33 to 24.46 mg.100 ml⁻¹. Similar results were obtained by Ding, et. al., [29] who found that this parameter was not significantly affected by the inclusion of monensin at a rate of 38 mg.kg⁻¹ into lambs diet, 25.69 to 26.58 mg.100 ml⁻¹.

3.2. Effect of interaction between level of concentrate feeding and addition of monensin on some biochemical blood parameter

The effect of interaction between level of concentrate and addition of monensin on blood parameters is shown in table 3. Results revealed that higher (P<0.05) BG concentration was detected in blood samples withdrawn from lambs fed higher level of concentrate (3%) with the addition of monensin, whereas, lower was detected in blood samples withdrawn from those fed higher level of concentrate (3%) without the addition of monensin. This increment in BG concentration may be due to the effect of monensin to increase the supply of glycolic precursors, thereby increasing the hepatic synthesis of glucose and consequently improving energy balance, with a high concentrate diet [5].

Higher BTP (P<0.05) concentration was detected in blood samples withdrawn from lambs fed the high level of concentrate with the addition of monensin (9.38 g.100 ml⁻¹) as compared with other samples. However, though insignificant, higher BTP concentration was detected in blood samples withdrawn from lambs fed the high level of concentrate without the addition of monensin (8.51 g.100 ml⁻¹) as compared with BTP concentration detected those withdrawn from lambs fed the lower level of concentrate without or with the addition of monensin (6.91 and 6.96 g.100 ml⁻¹). Similar result was reported by Drong [30]. The increase in BTP associated with diets containing a high level of concentrate with the addition of monensin may be attributed to the lower CP degradability of these diets [31]. The positive effect of low degradable protein can be simply explained by the reverse relationship between the rate of CP degradability and amino acid flow to the duodenum [32].

Results also showed that BUN concentrations were significantly decreased (P<0.05) by the addition of monensin regardless to concentrate level. But higher (P<0.05) concentration was associated with a high level of concentrate offered without monensin (9.81 mg.100 ml⁻¹) as compared with other diets. Nevertheless lower (P<0.05) BUN concentration was detected in blood samples withdrawn from lambs fed low level of concentrate with the addition of monensin (15.59 mg.100 ml⁻¹) (Table 15). Similar results were obtained by Xu, et. al., [33] where BUN concentration was significantly decreased (P<0.05) as affected by the interaction effect level of concentrate and between the addition of monensin.

Results also revealed that BTG was significantly (P<0.05) affected by the above interaction, where, as expected higher values were observed in blood samples withdrawn from lambs fed a high level of concentrate with the addition of monensin, whereas, those offered diet with the low level of concentrate without the addition of monensin recorded the lower concentration, values were 25.52 and 21.93 mg.100 ml⁻¹ respectively. Similar results were reported by Taghipoor, et. al., [34]. This may be due to the role of monensin in enhancing digestion.

3.3. Diurnal changes in some blood parameters

Diurnal changes in blood parameters as affected by factors studied in a current study are shown in table 4. Results indicated that higher (P<0.01) BG concentration was recorded in the samples withdrawn from the lambs before morning meal as compared with those withdrawn after 3 and 6 hrs of feeding. The concentration of BG was decreased (P<0.01) after 3 hrs of feeding. Then the reduction in the samples withdrawn after 6 hrs of morning feeding attained an additional decrease, values of 70.34, 66.50, and 60.73 mg.100 ml⁻¹, respectively. [35], obtained similar results, with the highest concentration of glucose obtained in the samples withdrawn before feeding which was then decreased to the lowest concentration at 6 hrs after feeding. [36], reported that the variations in the gluconeogenesis rate and synthesis of both insulin and glucagon were probably results for these fluctuations of glycemia during the day.

Regarding the diurnal changes of BTP concentrations, a higher (P<0.01) value was detected in blood samples withdrawn 3 hrs after morning meal (10.83 g.100 ml⁻¹), [37], reported that the increase in the amount of protein that reaches the small intestine and the use of amino acid was associated with increased BTP concentration. BTP concentration was then decreased (P<0.01) in samples withdrawn after 6 hrs of feeding (5.99 g.100 ml⁻¹) to return slightly increase after that to reach 6.86 g.100 ml⁻¹.
Results showed that higher (P<0.01) BUN concentration was recorded in samples withdrawn from lambs at 3 hrs after feeding, values were 18.61 mg.100 ml⁻¹. Then, it was decreased (P<0.01) to 16.58 mg.100 ml⁻¹ in samples withdrawn after 6 hrs of feeding. This pattern of changes in BUN concentration may indicate a gradual increase in the utilization rate of urea-N in the rumen. High BUN concentrations in lambs resulted from the high N intake and digestibility might be associated with high dietary CP content [16]. Therefore, BUN concentration can be a useful indicator of protein status in animals.

Results also showed that the lower value (P<0.01) of BTG concentration was detected in the blood samples withdrawn after 3 hrs of feeding (20.55 mg.100 ml⁻¹), as compared with those withdrawn before and 6 hrs thereafter. BTG concentration was increased (P<0.01) to 28.65 mg.100 ml⁻¹ in samples of blood withdrawn 6 hrs after morning feeding (28.65 mg.100 ml⁻¹). This pattern of changes was expected since it reflects dietary fat digestion and energy utilization.

**Table 1. Chemical composition of concentrate diet, its ingredients and wheat straw (%).**

| Ingredients        | ME MJ.100g⁻¹ |
|--------------------|--------------|
|                    | Ash | OM | CP  | CF  | EE  | NFE |      |
| Wheat bran         | 91.75 | 5.48 | 94.52 | 14.27 | 13.96 | 3.77 | 62.52 | 1.23 |
| Yellow corn        | 91.18 | 2.22 | 97.78 | 9.27  | 4.2  | 3.51 | 80.80 | 1.37 |
| Barley             | 91.78 | 5.65 | 94.35 | 10.16 | 6.71  | 1.99 | 75.49 | 1.27 |
| Soybean meal       | 91.93 | 7.87 | 92.03 | 45.48 | 3.75  | 1.83 | 39.35 | 1.18 |
| Urea               | -    | -   | -   | 287.5  | -    | -   | -    | -    |
| Concentrate        | 89.66 | 8.60 | 91.40 | 12.22 | 5.74  | 2.14 | 71.13 | 1.23** |
| Wheat straw        | 92.59 | 7.38 | 92.62 | 2.47  | 36.74 | 1.72 | 51.69 | 0.99** |

* 46% nitrogen × 6.25

ME values were estimated according to [9] equation with subsequent conversion from MJ. kg DM⁻¹ to MJ. 100 g DM⁻¹ in accordance with chemical composition based on percentage determinations:

**ME (MJ. kg DM⁻¹) = 0.012 CP +0.031 EE+0.005 CF +0.014 NFE

NaCl and mineral-vitamin mix were added to concentrate at rate of 1% for each. Urea was added at rate of 0.62% to ensure existence of a standard ratio of 1.34 g RDN. MJ⁻¹ of ME [10]. Level of RDN was estimated according to previous studies in which the ruminal effective degradability of protein fraction in the different ingredients of concentrate diet had been determined as follows: 80 and 60% for barley and yellow corn respectively [11], 70% for soybean meal [12] and 67% for wheat bran [13].

**Table 2. Main effect of level of concentrate feeding and addition of monensin on blood parameter (mean ± SE).**

| Blood parameter | Level of conc. % of BW | Monensin mg/kg conc. | P    |
|-----------------|------------------------|----------------------|------|
|                 | 2.5                    | 3                    | 0    | 30   | Conc. | Mon. |
| BG              | 66.02                  | 65.69                | 64.09 | 67.62 | NS    | NS   |
| mg/100 ml       | ±1.66                  | ±1.76                | ±1.73 | ±1.41 |       |      |
| BTP             | 6.94b                  | 8.84a                | 7.61b | 8.17a | **    | *    |
| g/100ml         | ±0.15                  | ±0.25                | ±0.29 | ±0.48 |       |      |
| BUN mg/100 ml   | 15.54b                 | 18.23a               | 18.15b | 15.62 | **    | **   |
| BTG             | 22.66b                 | 25.12a               | 23.33 | 24.46 |       |      |
| mg/100 ml       | ±0.57                  | ±0.26                | ±0.70 | ±0.49 |       | NS   |

Means in the same row with different superscripts are significantly different

** (P<0.01)  NS= Non significant

**Table 3. Effect of interaction between level of concentrate feeding and addition of monensin on blood parameter (mean ± SE).**

| Level of conc. % of BW¹ | 2.5% | 3% |
|-------------------------|------|----|
| Addition of monensin, mg/kg conc. | 0 | 30 | 0 | 30 | P |
| BG, mg/100 ml           | 66.44ab | 65.59ab | 61.74bc | 69.65a | * |
|                         | ±2.84   | ±2.19  | ±1.53   | ±1.32  |      |
| BTP, g/1000 ml          | 6.91c   | 6.96c  | 8.31b   | 9.38a  | *    |
|                         | ±0.24   | ±0.21  | ±0.15   | ±0.29  |      |
| BUN, mg/100 ml          | 16.50b  | 14.59a | 19.81a  | 16.65b | *    |
|                         | ±0.41   | ±0.39  | ±0.63   | ±0.19  |      |
| BTG, mg/100 ml          | 21.93c  | 23.39bc | 24.73ab | 25.52a | *    |
|                         | ±0.95   | ±0.53  | ±0.35   | ±0.33  | *    |

Means in the same row with different superscripts are significantly different , * (P<0.05)  NS= Non significant.
Table 4. Effect of time of sampling on blood parameter (mean ± SE).

| Blood parameter | Before feeding | After feeding, hrs | P     |
|-----------------|---------------|-------------------|-------|
|                 | 0 time        | 3                 | 6     |
| BG, mg/100 ml   | 70.34±        | 66.50±            | 60.73± |
|                 | ± 1.32        | ± 1.13            | ± 1.16 |
| BTP, g/100 ml   | 6.86±         | 10.83±            | 5.99±  |
|                 | ± 0.35        | ± 0.27            | ± 0.30 |
| BUN, mg/100 ml  | 15.44±        | 18.61±            | 16.58± |
|                 | ± 0.50        | ± 0.65            | ± 0.47 |
| BTG, mg/100 ml  | 22.50±        | 20.55±            | 28.65± |
|                 | ± 0.55        | ± 0.53            | ± 0.35 |

Means in the same row with different superscripts are significantly different
** *(P<0.01)*

Conclusions

The results suggest that the addition of monensin brought about additional benefits as evidenced by higher blood total protein and lower urea nitrogen concentrations. These may reflect improved utilization of diet by lambs.

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