

## Effects of Rumen Protected Arginine Supplementation on Growth Rate, Rumen Fermentation and Blood Biochemical of Awassi Lambs

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**ABSTRACT:** Twenty five Awassi male lambs weighing 24±0.92 kg and 3.5 months of old were used to investigate the effect of three levels of protected arginine (PA) (0.0, 5.0 and 7.0g PA/head/day) supplemented with cut twice a week (three days feeding, one day cut) on dry matter intake (DMI), live weight gain (LWG), feed conversion ratio (FCR) rumen fermentation rate and blood characteristics. Lambs were divided into five equal groups and fed randomly one of the following diets. The first group received the basal diet, composed of concentrate diet without PA which served as the control group (D1). The other tested groups were fed randomly on one of the following diets: D1 diet supplemented continuously with 5.0 or 7.0g PA /h/d (D2 and D3 respectively); D1 diet supplemented with cut twice a week 5 or 7g PA/h/d (D4 and D5 respectively). Total daily DMI are not significantly altered by level and methods of PA supplementation. The diets supplemented with PA showed best (P<0.05) daily LWG, total gain and FCR (D2, D3, D4 and D5) versus to control diet (D1). Ruminal pH and NH<sub>4</sub>-N were significantly (P<0.05) affected by levels of PA and methods of supplementation. Ruminal acetic acid are not affected, while propionic acid was significantly (P<0.05) increased with diets supplemented with PA. Blood parameters differed (P<0.05) significantly among treatments. Diets supplemented with PA caused higher (P<0.05) Urea-N, cholesterol, high density lipoprotein (HDL) and glutamate oxaloacetate transaminase (GOT) and lower (P<0.05) low density lipoprotein (LDL) and glutamate pyruvate transaminase (GPT) as compared with control diet (D1). In conclusion, supplementation of Awassi lambs diets with PA improved LWG, FCR, rumen fermentation rate and some blood parameters.

**Key Words:** arginine, growth rate, rumen fermentation, blood parameters, lambs.

### INTRODUCTION

Arginine is a basic amino acid known for its stimulatory effect on Luteinizing Hormone and Growth Hormone release in sheep (Recabarren et al., 1996; Davenport et al., 1990) and goats (Basiouni et al., 1999). The changes in arginine metabolism and requirements in ruminants (Ball et al., 2007) who observed no specific amino acid requirements for ruminants such as growing beef (NRC, 2000) or dairy (NRC, 2001) cattle because these animals were obtain enough amino acids from milk during suckling period (Williams and Hewitt, 1979), and when the rumen is developed, ruminants receive enough amino acids from the combination of diet and ruminal microbial amino acid synthesis. However, in calf body protein, arginine intake was found to be only 60% of the estimated metabolic requirement (NRC, 2001). Furthermore, in milk replacer-fed calves, weaning weight and plasma arginine concentrations were greater when calves received an arginine-supplemented formula (Fligger, et al., 1997). The administration of arginine into the abomasum of growing calves in conjunction with ammonium acetate improved nitrogen retention and lowered plasma ammonia concentrations relative to ammonium acetate administration alone (Koenig, et al., 1982). The estimated dietary arginine requirement of 8.5 g/d in a 500 kg cow (NRC, 2001) is very low compared with the dietary requirements of other growing mammals. However, because this requirement

was based only on amino acid composition of body protein, it does not reflect the entire need for arginine to support urea cycle function; therefore, the requirement may be underestimated. The ruminant liver has detectable activity of all of the urea cycle enzymes (Chalupa, et al., 1970). Ruminants extensively utilize nitrogen recycling; blood urea is secreted into the gastrointestinal tract, where it is used by the rumen microflora to resynthesize amino acids (Marini and Van Amburgh, 2003). Therefore, it would be predicted that increasing arginine supply to the liver would increase the efficiency of incorporation of ammonia into carbamoyl phosphate and urea synthesis, which may ameliorate effects of ammonia on AAs catabolism. Arginine infusion into the abomasum of beef heifers resulted in increased body N retention and decreased arterial concentrations of AA (Davenport et al., 1990), responses to arginine supplementation is partly related to changes in liver metabolism. Up to date, there is no research has specifically conducted, to examine the effects of super physiological intakes of arginine on growth rate, rumen fermentations and blood parameters in sheep. Our objectives were to investigate the effect of levels and methods of supplementation protected arginine (PA) on daily intake, live weight gain, rumen fermentation and blood characteristics in growing Awassi lambs.

## MATERIAL and METHODS

### Protected arginine (PA)

To protect arginine from rumen degradation, formaldehyde treatment of arginine was achieved by spraying formaldehyde (4%) solution into the arginine at the ratio of 10 ml /100 g arginine DM, equivalent to 1 g formaldehyde per 100 g of air dry arginine (Preston and Leng 1985 and Hassan, et. al., 1990), using simple sprayer. The treated arginine was then mixed well and packed into polyethylene sacs which were tightly closed and left at room temperature for 3 days and were shaken occasionally, then all sacs were opened and the treated arginine was exposed to air to remove the excess formaldehyde before mixing with the other ingredients.

### Animals and Diets

Twenty five male Awassi lambs, aged 3.5 months and averaged  $24 \pm 0.92$  kg live body weight were used. Lambs were divided into five similar groups (Five lambs for each) and assigned to the five experimental diets. Animals were individually housed at the breeding and improvement of sheep and goats station/Ministry of Agriculture/ in the area of Ekrkov (25 km) west of Baghdad for the period from May 2009 to August 2009. The effect of three levels (0.0, 5.0 and 7.0 g PA/h/d) and two methods (contentious and cut twice a week) of protected arginine supplementation were investigated. The first group received the basal diet, composed of concentrate diet without PA which served as the control group (D1). The other tested groups were fed randomly on one of the following diets: D1 diet supplemented continuously with 5.0 or 7.0 g PA /h/d ( D2 and D3 respectively ); D1 diet supplemented with 5.0 or 7.0 g PA/h/d with cut twice a week (three days feeding one day cut, D4 and D5 respectively). The formulation and chemical composition of concentrate diet and its contents are presented in Table 1. The diets were gradually introduced to the lambs over a period of 3 weeks before the start of experiment, during this time

all animals treated for tapeworms and other helminthes. Animals were gradually introduced to the level of 3 % of live body weight of concentrate diet, alfalfa hay was offered ad libitum and containing (% of dry matter, DM basis): 95% DM, 92% organic matter (OM), 2.25% nitrogen(N), 10.2% metabolisable energy (ME), 46% nutria detergent fibre (NDF), 30% acid detergent fibre (ADF) and 18% lignin. Animals were fed once daily at 09.00 h and had free access to fresh water. Live body weight (LBW) was recorded once weekly to the nearest 0.25 kg. Feed intake was determined as the difference between feed offered and refused.

### Rumen fermentation characteristics

At the end of the experiment and within one day, rumen fluid samples were withdrawn from half of the lambs 12 hr post morning feeding to study rumen fermentation characteristics through the determination of the ruminal pH,  $\text{NH}_3\text{-N}$  and volatile fatty acid (VFA) concentrations. The lambs were sampled using the smooth stomach tube which connected to 50 ml syringe as described by Saeed (2008). Rumen fluid was immediately measured for pH. Samples were then filtered through four layers of cheesecloth to discard the solid unfermented particles. Then 10 ml subsamples were preserved by addition of 0.2 ml sulfuric acid 50% to kill bacterial action and capture ammonia, and stored at  $-20\text{C}^0$ . Until subsequent analysis (Kazemi-Bonchenari, et. al., 2010). Just before analysis, frozen strained rumen fluid samples were thawed at room temperature and shaken, then the contents were transferred into glass tubes and centrifuged (80-1, China) at  $4,000 \times g$  for 20 minutes. The supernatant was analyzed for ruminal ammonia-N by the method of steam distillation with MgO using a Kjeltec (Gerhardt-Germany) distillate unit (AOAC, 1990), total VFA (TVFA) by the method of Margham (1942) and the molar percentages of individual VFA (HPLC, model RF-10AXmugiL, Shimadzu, Japan) according to Zinn and Owens (1986).

Table1. Formulation and chemical composition of concentrate diet and its contents (% of DM basis)

Chemical composition	Ingredients %	DM %	OM%	CP%	EE%	CF%	NFE%	RDN <sup>*</sup> %	ME <sup>**</sup> MJ/Kg DM
Barley	34	31.19	29.38	3.77	0.54	2.72	22.34	80	3.88
Yellow corn	20	18.09	17.0	1.86	0.78	0.62	13.74	60	2.35
Wheat bran	30	27.21	25.49	3.57	1.13	4.38	16.41	67	3.29
Soybean meal	14	12.59	11.63	5.62	0.27	0.71	5.04	70	1.50
Salt	1.3	-	-	-	-	-	-	-	-
CaCO <sub>3</sub>	0.7	-	-	-	-	-	-	-	-
Concentrate	100	91.07	83.50	14.82	2.72	8.43	57.53	69.1	11.10

\* RDN, rumen degradable nitrogen was estimated according to published effective degradability of ingredients as follows: barley 80% and yellow corn 60% (Humady, 1988), wheat bran 67% (Paya, et al., 2008), SBM 70% and Alfalfa hay 72% (Hassan and AL-Sultan, 1995).

\*\* Calculated according to MAFF (1975).

### Blood sampling and preservation

At the of feeding trail, five ml of blood samples were collected from the experimental animals in each

treatment (3 Lambs) by jugular vein puncture just 12 hr post feeding to determine serum urea nitrogen (SUN), serum cholesterol (SC), glutamate oxaloacetate

transaminase (GOT), glutamate pyruvate transaminase (GPT), low density lipoprotein (LDL) and high density lipoprotein (HDL). The blood samples were centrifuged at 4000 r.p.m. for 20 minutes, and blood serum was separated and preserved in clean and sterile a plastic tube which was stored at -20 C° for biochemical assay. Mean serum concentration were calculated for each animal within each treatment group. SUN was measured photo metrically in the serum fraction using a Digital Spectrophotometer (PD-303). APEL-JAPAN, the kit that used to analyze SUN was marked as (Urea-kit S180, France) and according to the method of Coulombe and Faveran (1963). The concentration of SC was determined calorimetrically by using commercial kits according to Schmidt-Nielsen (1964). The kit that used to analyze total protein was marked as (BIOLABO REAGENTS 02160, France). Serum GOT and GPT activities were determined calorimetrically by using commercial kits according to Armstrong and Carr (1964).

#### Chemical analysis

Proximate chemical analysis of alfalfa hay and concentrate samples in triplicate per each determination was carried out for dry matter (DM), crude protein (CP), ether extract (EE), crude fiber (CF) and ash content according to the A.O.A.C. (2001). The nitrogen free extract (NFE) was calculated by subtracting the summation percentages of CP, EE, CF and ash from one hundred. Alfalfa hay samples was analyzed according to Goering and Van Soest (1970) to determine neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL). Hemi-cellulose and cellulose were determined by difference.

#### Statistical analysis

Data was statistically analyzed using Completely Randomized Design Model (CRD) procedure by (SAS, 2001). Duncan's multiple range test was used to determine the significance of differences between treatments means Duncan (1955). Analysis of variance was carried out on all data. The treatment was partitioned into main effects and their interaction.

### RESULTS and DISCUSSION

#### Daily intake

In general, the lambs consumed all the concentrate diet offered (3% of LBW). Daily intake of total DM, N, ME, RDN, UDN, initial and final live body weight (FLBW), total live weight gain (TLWG), daily live weight gain (DLWG) and feed conversion ratio (FCR) as affected by level of PA, supplemented continuously or with cutting interval are presented in Table (2). The DM intake (g/d) of concentrate diets and alfalfa hay supplemented with PA was slightly different than control diet (D1), but these differences were not statistically significant. As well as, total daily intake of DM, N, ME and RDN are not significantly altered by

level or methods of PA supplementation. This result is in agreement with other studies (Christensen, et. al., 1993; Devant, et. al., 2000; Bohnert, et. al., 2002b and Braud, 2005) and it seems that daily RDN and UDN intake of lambs in all treatments (1.5 g RDN/MJ of ME and 11g UDN/day) are often completely met through their allowances of concentrate diets and above ARC (1984) recommendation, hence, there is no need to alfalfa hay which is rich roughage in both nitrogenous fractions to derive more of these nutrients. Kellaway and Leibholz (1983) reported that when RDN is non-limiting, fed at moderate levels have negligible effects on roughage intake. Even though, ruminants require some source of forage to ensure normal rumen function (Mulligan, et. al., 2001). In contrast, other study observed that high AAs of UDN diet stimulate higher DMI as compared to low UDN diet (Nisa, et. al., 2008 ). The contrariety may attributed to RDN:UDN ratios. A constant ratio was used in this study or may be attributed to level of feeding, offering concentrate at level of 3 % of body weight may forbad the promising effect of high RDN : UDN ratio from being appear. The positive effect of high AAs by pass fraction have been observed by other studies (Chaturvedi and Walli, 2001).

#### Live weight gain

As well as, Table 2 shown that FLBW, TLWG and DLWG of lambs fed diets supplemented with PA either continuously (D2 and D3) or with cut intervals (D4 and D5) were increased significantly ( $P<0.05$ ) as compared with those fed the control diets (D1). However, differences were not statistically significant across D2, D3, D4 and D5 diets. Moreover, interaction between level and methods of PA supplementation was not significant. Similar findings were observed by many other reports (Zerbini and Polan, 1985; Firkins, et. al., 1986; Braud, 2005). The level of RDN in a current study may be higher than exactly required by lambs as evidenced by increasing ruminal  $\text{NH}_3\text{-N}$  and SUN (Tables 3 and 4). Nisa, et. al., (2008) reported that higher level of RDN in a ruminant ration causes excessive ruminal  $\text{NH}_3$  production which ultimately results in an increased BUN. Excess ammonia is transported to the liver and converted to urea N that is excreted in the urine or recycled back to the rumen (Leng and Nolan, 1984). Khampa, et. al., (2003) observed that 12% of RDP: DOMI was beneficial in maximizing performance, in a current study this ratio was 13.23 % in concentrate diet. In a current study, concentrate constitutes about 60% of total rations. More importantly, the level of concentrate feeding (3% of LBW) may lead to cover the effect of RDN level. Ammonia is probably the most important source of N for growth of ruminal bacteria (Allison, 1969). In a current study concentrates and hay are offered sequentially once a day. Whereas, UDP may be better suited to less frequent supplementation because of its delayed degradation (Bohnert, et. al., 1998) compared with RDP. This could result in increased N recycling to

the gut (due to lower ruminal ammonia levels) and decreased urinary N excretion. On the other hand, Weiss, (2002) declared that ruminant do not require RDP but rumen microorganism do. Many studies referred to the beneficial effect of UDN supplementation (Hassan and Bryant, 1986; Habib, et. al., 2001; Chaturvedi and Walli, 2001; Flis and Wattiaux, 2005; Gulati, et. al., 2005; Yörük, et. al., 2006; Atkinson, et. al., 2006; Nisa, et. al., 2008). UDP supplementation improves the quality and quantity of

AA reaching the small intestine for absorption (Merchen and Titgemeyer, 1992). This change in AA profile could cause a growth response to occur (Tomlinson, et. al., 1997). This may lead to improve N balance, Pattanaik, et. al., (2003) reported higher N retention in calves fed a low degradable protein diet than in those fed high degradable protein diet. Similar findings were reported by Paengkoum, et. al., (2004/6) who described that N retention decreased linearly as the dietary UDN level decreased.

Table 2. Performance of Awassi lambs as affected by supplementation of protected arginine (PA) to the concentrate ration.

Methods of PA supplementation	Control	Continuous supplementations			Cut interval supplementation	
Levels of PA (g/h/d)	0	5	7	5	7	
Diet no	D1	D2	D3	D4	D5	
Daily intake g/d						
Concentrate	871±22	893±23	901±19	882±20	902±21	
Alfalfa Hay (DM)	690±33	698±30	700±23	711±32	740±22	
Total DM (TDM)	1561±56	1591±54	1601±44	1593±58	1642±64	
Total nitrogen (TN)	36.1±0.90	35.6±0.99	36.9±0.86	36.8±0.78	37.9±0.96	
ME (MJ)	16.96±0.33	17.02±0.43	17.25±0.27	17.04±0.31	17.5±0.29	
RDN	25.2±0.66	24.9±0.19	25.8±0.14	25.7±0.17	26.5±0.12	
RDN g/MJ of ME	1.49±0.04	1.46±0.09	1.49±0.08	1.5±0.05	1.5±0.5	
UDN	10.9±0.62	10.7±0.26	11.1±0.28	11.1±0.33	11.4±0.17	
Initial Live weight (Kg)	24.82±0.87	24.84±0.91	24.82±1.05	24.72±1.01	24.84±1.13	
Final Live weight (Kg)	34.92±0.83 <sup>b</sup>	37.18±1.15 <sup>a</sup>	37.0±0.81 <sup>a</sup>	37.38±1.01 <sup>a</sup>	38.20±1.45 <sup>a</sup>	
Total gain (Kg)	10.1±0.53 <sup>b</sup>	12.3±0.78 <sup>b</sup>	12.2±0.50 <sup>b</sup>	12.7±1.06 <sup>a</sup>	13.4±0.96 <sup>a</sup>	
Live weight (LWG, g/d)	120±6.35 <sup>b</sup>	147±9.26 <sup>a</sup>	145±6.15 <sup>a</sup>	150±12.7 <sup>a</sup>	159±11.49 <sup>a</sup>	
Feed conservation ratio g TDM/ g LWG	13.0±0.49 <sup>a</sup>	10.8±0.31 <sup>b</sup>	11.0±0.34 <sup>b</sup>	10.6±0.49 <sup>bc</sup>	10.3±0.35 <sup>c</sup>	

<sup>abc</sup> Means with the different superscripts within row are significantly (P<0.05) different

Table 3. Rumen fermentation of Awassi lambs as affected by supplementation of protected arginine (PA) to the concentrate ration

Methods of PA supplementation	Control	Continuous supplementations			Cut interval supplementation	
Levels of PA (g/h/d)	0	5	7	5	7	
Diet no	D1	D2	D3	D4	D5	
Items						
pH	6.01±0.00 <sup>c</sup>	6.62±0.00	7.38±0.05	6.54±0.03	5.87±0.00	
Ammonia nitrogen mg/100 ml	90.5±0.50 <sup>b</sup>	698±30	53.5±27	53.5±27	67.0±0.33	
Total volatile fatty acids mmol/l						
Acetic acid molar proportion (%)	52.5±0.65	52.1±0.77	54.4±0.62	53.9±29	52.4±0.21	
Propionic acid molar proportion (%)	28.0±0.31 <sup>b</sup>	17.02±0.43 <sup>b</sup>	34.2±0.44 <sup>b</sup>	35.1±0.23 <sup>b</sup>	37.3±0.04 <sup>a</sup>	
Butyric acid molar proportion (%)	19.5±0.95 <sup>a</sup>	24.9±0.19 <sup>b</sup>	11.3±0.27 <sup>b</sup>	11.0±0.35 <sup>b</sup>	10.3±0.04 <sup>b</sup>	

<sup>abc</sup> Means with the different superscripts within row are significantly (P<0.05) different

#### Feed conversion ratio

As well as, feed conversion ratios (Table 2) values are significantly affected by levels of PA supplementation. The diets supplemented with PA showed best feed conversion ratio (D2, D3, D4 and D5) versus to control diet (D1). However, diets D4 and D5 showed better improvement (P<0.05) in FCR as compared with diets D2 and D3. Interaction between level and methods of PA supplementation was statistically significant (P<0.05). This result is agree with findings of Khan, et. al., (2000) and Abdullah and

Awawdeh (2004) who reported that formaldehyde treatment of arginine led to improve FCR. This improvement was attributed to increase bypass arginine to small intestine (SI), where utilization of AAs and glucose absorbed via SI instead of its extensive fermentation in the rumen is more efficient (Meissner, et. al., 1996). Therefore, it is expected that feeding high protected AAs may increase the efficiency of utilization of diet.

### Rumen fermentation parameters

Rumen fermentation parameters as affected by levels of PA and methods of supplementation and their interaction are presented in Table 3. Ruminant pH was significantly ( $P<0.05$ ) affected by levels of PA and methods of supplementation. where, higher ( $P<0.05$ ) value of pH was observed when lambs fed diet supplemented with high levels of PA continuously (D3), however, lower value was observed with high level of PA with cut interval (D5), this result pushes to believe that ruminal pH may be affected by quality of protein but not its quantity (Driedger, et. al., 1998). Moreover, ruminal pH was significantly affected ( $P<0.05$ ) by the interaction between levels and methods of PA supplementation. This results pushes to believe that ruminal pH may be affected by quality and quantity of AAs and methods of supplementation (Driedger, et. al., 1998). It is also shown that ruminal  $\text{NH}_3\text{-N}$  was significantly ( $P<0.01$ ) affected by levels of PA and methods of supplementation. Higher ( $P<0.01$ ) ruminal  $\text{NH}_3\text{-N}$  concentrations are recorded due to feeding diets supplemented continuously with PA (D2 and D3) as compared with those fed control diet, D4 and D5, whereas, lower ( $P<0.01$ ) ruminal  $\text{NH}_3\text{-N}$  concentrations are recorded due to feeding diets supplemented with PA with intervals cut (D4 and D5). Increased ruminal  $\text{NH}_3\text{-N}$  concentration due to increasing RDN fraction were previously indicated by several studies (Mathieu, et. al., 1996; Fievez, et. al., 2001; De Boever, et. al., 2005; Gressley and Armentano, 2007 and Nisa, et. al., 2008). The levels of RDN in current study were formulated to be similar across treatments. Thus the lower ammonia concentration may refer to higher utilization by rumen microbes (Devant, et. al., 2000; Lascano and Heinrichs, 2009). Regarding TVFA, statistical analysis revealed that TVFA concentrations were not significantly affected by level of PA and methods of

supplementation, Similar results was obtained by Bargo and Rearte (1998), Who reported that total VFA concentration in ruminal fluid was not affected by feeding high and low ruminally degradable protein sources. This result disagrees with those found by Köster, et. al., (1996) who reported that ruminal TVFA concentrations were increased ( $P<0.01$ ) in response to RDP supplementation, While, Brossard, et. al., (2003) observed that TVFA were decreased ( $P<0.001$ ) due to increasing degradability of dietary protein. In general high level of TVFA observed across treatments may attributed to formation of organic acids in the rumen accompanied with feeding high level of concentrate, Galip (2006) reported that TVFA concentrations significantly increased in rumen after feeding. Similar to ruminal TVFA, acetic acid molar proportion was not significantly affected by level and methods of PA supplementation, while, higher ( $P<0.01$ ) propionic acid was achieved by lambs fed diets supplemented with PA (D2, D3, D4 and D5) as compared with control diet (D1). However, higher propionic acid was achieved by lambs fed diets supplemented with 7.5g PA with interval cut (D5). Brossard, et. al., (2003) observed that TVFA concentration ( $P<0.001$ ), acetate ( $P<0.001$ ) and propionate ( $P<0.01$ ) proportions in the rumen decreased due to increasing degradability, while the butyrate proportion increased ( $P<0.001$ ). In a current study, there is a significant ( $P<0.01$ ) decrease in ruminal butyric acid due to feeding diets supplemented with PA, as compared with control diet (D1). However, this significance was not seen among D2, D3, D4 and D5 diets. It has been noticed that marked changes in the molar proportions of the concentrations of ruminal individual VFA can be induced in response to the inclusion of arginine of UDN sources differed in degradability rate (Sutton, et. al., 2003).

Table 4. Blood parameters of Awassi lambs as affected by supplementation of protected arginine (PA) to the concentrate ration

Methods of PA supplementation	Control	Continuous supplementations		Cut interval supplementation	
Levels of PA (g/h/d)	0	5	7	5	7
Diet no	D1	D2	D3	D4	D5
Items					
pH	50.05±0.50 <sup>b</sup>	60.7±0.17 <sup>a</sup>	59.2±0.11 <sup>a</sup>	53.4±0.12 <sup>b</sup>	50.4±0.07 <sup>b</sup>
Ammonia nitrogen mg/100 ml	102±0.00 <sup>d</sup>	130±0.26 <sup>b</sup>	127±0.14 <sup>b</sup>	156±0.39 <sup>a</sup>	122±0.27 <sup>b</sup>
Total volatile fatty acids mmol/l	47.0±0.00 <sup>a</sup>	27.9±0.09 <sup>b</sup>	23.7±0.08 <sup>c</sup>	20.4±0.32 <sup>d</sup>	21.4±0.38 <sup>d</sup>
Acetic acid molar proportion (%)	18.5±0.50 <sup>c</sup>	21.7±0.24 <sup>b</sup>	22.4±0.05 <sup>b</sup>	25.4±0.13 <sup>a</sup>	25.1±0.08 <sup>a</sup>
Propionic acid molar proportion (%)	10.5±0.50 <sup>a</sup>	6.9±0.13 <sup>b</sup>	8.4±0.10 <sup>b</sup>	7.4±0.09 <sup>b</sup>	8.5±0.18 <sup>b</sup>
Butyric acid molar proportion (%)	19.0±0.00 <sup>c</sup>	49.2±0.07 <sup>a</sup>	34.8±0.12 <sup>b</sup>	32.6±0.11 <sup>b</sup>	32.1±0.05 <sup>b</sup>

<sup>abc</sup> Means with the different superscripts within row are significantly ( $P<0.05$ ) different

### Blood parameters

Blood parameters as affected by levels of PA supplemented continuously or with cut intervals and their interaction are presented in Table 4. Urea-N were pointed out to, ( $P<0.05$ ) differences among treatments in favor of diets supplemented continuously with PA (D2 and D3) which recorded the highest ( $P<0.01$ ) urea-

N concentration in comparison with the other diets (D1, D4 and D5). However, differences between D4, D5 and control group were not statistically significant. The urea-N level ranged from 50.4 in D5 to 60.7 mg/dl in D2. Rakha, (1985) reported that the normal urea-N level in sheep and goats was ranged from 8 to 40 mg/dl. Changes in serum urea would reflect changes in

criminal ammonia-N concentration (Fouda, 2008). Such results may favored urea as a good soluble degradable NPN source which led to enrich animal rumen media with an abundance N released. Thus the excess  $\text{NH}_3\text{-N}$  which was more than the ability of microflora to synthesis to microbial protein is being withdrawn via the portal vein to animal livers and in turn raised urea concentrations in serum blood of experimental animals (Hassan and Hassan, 2010b; Hassan et al., 2009b). Our results were in accordance with those of Lewis (1957) who demonstrated that increasing the concentration of plasma urea after feeding was caused by the increasing of ruminal ammonia. Moreover, blood urea N analyses can be used as a signal red to point out potential problem in the feeding program. The BUN level in excess of 18 to 20 mg/dl can be associated with lower reproductive performance, higher feed costs, health problems, and poor production (Hansen, 2003). Finally, the obtained results indicated that the PA supplementation with interval cut had no adverse effect on liver or kidney function. Similar results were reported by Hassan and Hassan (2009 and 2010) when lambs fed diets supplemented with different levels of rumen degradable and undegradable nitrogen respectively. Results also indicated that the diets supplemented with PA were significantly ( $P<0.05$ ) increase cholesterol and GOT, and significantly reduced ( $p<0.05$ ) LDL, HDL and GPT as compared with control diet (D1). However, differences were statistically significant ( $P<0.05$ ) among D2, D3, D4 and D5 diets. Higher ( $P<0.01$ ) levels of cholesterol, HDL and GOT are recorded due to feeding diets D4, D4 and D2 supplemented with PA respectively as compared with other diets, whereas, lower ( $P<0.01$ ) levels of LDL and GPT are recorded due to feeding diets D4 and D2 supplemented with PA respectively. Except, that control diet D1 recorded higher ( $P<0.01$ ) levels of LDL as compared with D2, D3, D4 and D5 diets. Cholesterol values obtained in the present study were within the normal ranges obtained by Hassan et al., (2009 and 2010) using Awassi and Karadi lambs. GOT and GPT assessed in blood serum, as an indicators of liver function showed a significant differences among different experimental groups.

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