

(Original Article)



The Effect of Enzymes Mixture Supplementation on Productive Performance of Ewes and Their Lamb

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Abstract

The objective of this study was to investigate the impact of supplement of Exogenous enzymes mixture for local ewes and rams on nutrient digestibility coefficients, rumen fermentation parameter, milk production, some blood metabolites, and lambs' performance. Twenty-one pregnant ewes during the last two months of pregnancy randomly distributed into three different groups (seven animals each), depending on their mean live body weight and season of production. The 1st group was a control or basal diet without enzyme supplement, the 2nd (T1) and 3rd (T2) groups were fed a basal diet with a supplement of 30 and 60 g enzymes mixture /100 kg concentrate feed mixture (CFM), respectively. The experimental period lasted for three months after lambing. Results showed that the enzyme mixture supplementation did not affect the digestibility and nutritional values of diets. The mean milk yield during the lactation period was higher ($P < 0.05$) in T1 than in other groups. Milk fat was higher ($P < 0.05$) with T1 than with T2. Ruminal total volatile fatty acids (TVFA's) were higher in T1 than in T2 and the controls. Also, the concentration of ammonia nitrogen (NH₃-N) was higher ($P < 0.05$) in the treated groups than control animals. The total protein and globulin were increased ($P < 0.05$) in (T1) compared with T2 and control groups. The average daily gain of lambs was ($P < 0.05$) greater in T1 than in the control group. It could be concluded that supplement of enzymes mixture to ewes' diet during the last two months of pregnancy and lactating period, particularly at a rate of 30g/100 kg of CFM improved milk yield, milk fat, some blood metabolites, rumen fermentation, and the weight of newborn lambs at weaning.

Keywords: *Enzyme mixture, Digestibility, Milk production, Pregnancy.*

Introduction

It is known that ruminants depend on the accompanying microbial communities and cannot synthesis enough necessary fibrolytic enzymes for the breakdown of lignocelluloses, the use of feed substance and enzymes in animals' nutrition has recently attracted the interest of nutritionists. These days using of

exogenous fibrolytic enzymes in ruminant diets has been grown because of their ability to increase feed digestion *in vitro* and *in vivo* (Yang *et al.*, 1999, Bowman, 2002, Kung *et al.*, 2002).

Many studies indicated that increased feed intake due to fibrolytic enzymes supplementation, which may partially be attributable to the diet's improved palatability due to the sugars produced by pre-ingestive fiber breakdown. However, the effects of post-ingestive enzymes, such as an increase in the efficiency or level of digestion (Gado and Salem, 2008; Krueger and Adesogan, 2008). External enzymes added to ruminant diets have been shown to have beneficial impacts on developing cattle and suckling dairy cows (Gado *et al.*, 2009). The addition of fibrolytic enzymes to the lactating cattle rations increased feed intake and improved milk yield by about 2-25% (Tricarico *et al.*, 2005), improved the energy balance of transition dairy cows (DeFrain *et al.*, 2005) and an increased in the milk yield of young ruminants (Titi and Lubbadeh., 2004). Gado *et al.* (2009) found that giving external enzymes to dairy cows fed on total mixed rations enhanced ($P < 0.05$) rumen microbial N synthesis, total volatile fatty acids (TVFA's) concentrations, feed intake, and digestibility of organic matter. Gomaa *et al.* (2012) stated that ruminal ammonia-N, (TVFA's), plasma total protein, TDN%, nutrients digestibility, and plasma total protein values were increased ($P < 0.05$) in sheep with the added enzymes to rice straw-based rations.

The beneficial results of exogenous enzymes supplementation rely upon several variables, including the content of the diet, the kind of enzyme preparation, the enzyme's stability, the specific enzymatic action, the quantity of enzyme supplied, and the application technique (Yang *et al.*, 2000 and Morgavi *et al.* 2001). However, some investigators found that sheep weight gain, feed conversion, digestibility, and rumen kinetics were not affected by enzyme treatment (Giraldo *et al.* 2009; Bueno *et al.*, 2013; Torres *et al.* 2013). The aims of the current study were investigating the impact of enzymes mixture supplementation to pregnant ewes on ruminal fermentation, nutrient digestibility, ewes and their lamb performance.

Materials and Methods

Animals, diets and management

This experiment was conducted at the Experimental Farm of Animal Production Department, Faculty of Agriculture, Al- Azhar University, Assiut branch, Egypt. The experiment started from October 2021 to March 2022. Twenty-one pregnant local ewes at the last two months of pregnancy (1.5- 2 years old with an average live body weight of 40.50 ± 2.20 kg) in the second and third seasons of production were randomly distributed into three equal groups. In all groups, the average beginning weights were similar. Ewes in each group were fed individually. The experimental period included two months of pregnancy and continued three months after lambing. The enzymes mixture (Cairo Biopharm®, for veterinary products and feed additives, the Egyptian Factory, Egypt). Each

Kg of enzymes mixture consists of protease 25000 IU, xylanase 20000 IU, phytase 20000 IU, β glucanase 100000 IU, amylase 100000 IU, hemicellulase 35000IU, cellulase 25000 IU, lipase 25000, sodium metabisulfite 20000 mg, pectinase 10000 IU and papain 5000 IU.

The experimental groups were as follows, the 1st group was control or basal diet (without additives), while the 2nd and 3rd groups were fed the basal diet with a supplement of the enzymes' mixture by 30 and 60 g /100 kg CFM, respectively. All animals covered 60% of their requirements as concentrate feed mixture, while roughage was added *ad libitum*. Every two weeks, the amount of the concentrate mixture was changed to account for variations in the ewes' body weight (NRC, 1985). Fresh water and licks of minerals and vitamins were available free of choice. The CFM was given once daily at 7.00 a.m., while wheat straw was fed at 11.00 a.m., throughout the trial period, the feed orts was weighed daily, and actual feed consumption was calculated. Ingredients and chemical composition of CFM and wheat straw were presented in Table (1). The chemical analysis of feeds was analyzed using the procedures of the Association of Official Analytical Chemists AOAC (2005). Lambs were weighed at birth and biweekly after that up to weaning for three months of age to calculate the average daily gain body weight gain.

Digestion trials

The digestibility trials were conducted using nine rams (two years old and weighed about (55 ± 2.67) kg). The rams were kept individually in metabolic cages with free access to water. The study lasted for 21 days, the first 14 days were the preliminary period and the next 7 days were used to collect faeces and feeds residues. Rams were divided into three groups, the first group was a control, while the second (T1) and third (T2) groups the animals received 30 and 60 g enzyme mixture /100 kg CFM, respectively. According to the NRC (1985), the rations were formulated to meet the requirements of rams. All rams of all groups were fed 60% of their requirements as a concentrate feed mixture while wheat straw was added *ad libitum*. The daily feed consumption of concentrate feed mixture and roughage was estimated by subtracting residual feed from the provided one. Dietary feed samples were collected, mixed, and grinding through a 1 mm screen before being kept for chemical analysis. Every day, feces were collected and 10% of its weight was taken. The fecal samples from each ram were composited and dried at 60°C for 24hr before grinding through a 1mm mill screen for chemical analysis. Using AOAC (2005) methods, chemical analysis of feed samples and feces were done. The feeding value, in terms of total digestible nutrients (TDN) and digestible crude protein (DCP), was determined using chemical analyses of the ingredients utilized and the apparent digestibility rates of the various nutrients in the diets on the basis of McDonald *et al.* (1988).

Blood sampling

Blood samples were collected monthly at the end of suckling period by taking 10 ml blood from the jugular vein of each lamb, at 6 h after the morning

feeding in glass tubes continuing anticoagulant (EDTA). Samples of blood were immediately centrifuged at 4000 rpm for 15 min. and the plasma was kept at -20 °C until analysis. Plasma was used to measure the total protein (g/dl), albumin (g/dl), glucose (g/dl), cholesterol (mg/dl), and triglycerides (mg/dl) using commercial kits produced by Spectrum Chemical Company using spectrophotometer method. According to Titaz (1976), plasma total protein and albumin were determined, while globulin values were determined by subtracting albumin values from total protein values. Triglyceride levels in plasma were determined according to Fassati and Principe, (1982). Plasma glucose and plasma cholesterol were measured in accordance with Titaz (1976) and Younge (2001), respectively.

Milk yield

Using the lamb's suckling technique, daily milk yield (g/day) for each ewe was measured after the 15th day from lambing to the weaning period of the lactation period (90 days) as described by Ashmawy (1980) and Morsy (2002). This technique was used twice with each ewe per week. Individual samples of milk were taken from all ewes during the suckling period. The samples were gathered early in the day and afternoon and then mixed for analysis using a 50 ml milk sample to determine milk fat, protein, lactose, total solids, salts, and not solids fat using a milk analyzer (Lactoscan MCCW. 5365).

Rumen liquor characteristics

Samples of rumen fluids were obtained on two consecutive days after the digestibility trial's collection period ended using a stomach tube. Samples of rumen fluid were collected before morning feeding (zero time) and then after 4hr. of feeding. Four layers of cheesecloth were used to filter samples of rumen fluid. A digital pH meter (Beckman, model 45, USA) was used to measure the rumen pH of the filtrate portion. Strained samples of rumen fluid were acidified with 0.1 N hydrochloric acid and 2-3 drops of formalin or formaldehyde to inhibit microbial growth, and then the samples were kept frozen at -20°C for determination of TVFA's and ammonia-N concentration. Using a Markham micro distillation apparatus, the concentration of TVFAs was measured using the steam distillation method (Wanner, 1964). The concentration of ammonia -N in rumen fluid was measured using the kjeldahle distillation method AOAC (2000).

Statistical analysis

General linear model (G.L.M) statistical analysis was used to examine the data. procedure of S.A.S (2001) program, version 8.2. Differences between treatment for nutrient digestibility, nutritive values, feed intake, blood parameters, milk production, milk composition, and lamb performance. were evaluated by one-way ANOVA. Using the Duncan Multiple Range Test, the significant differences between treatment means were determined. (Steel and Torrie, 1980). The data are presented as means and \pm SE. P. values less than ($P < 0.05$) were considered significant. The next model was applied: $Y_{ij} = \mu + T_i + E_{ij}$

The data for the rumen liquid properties were examined using the following statistical model.

$$Y_{ijk} = \mu + T_i + M_j + (TM)_{j} + E_{ijk}$$

Where, Y_{ij} = experimental observation, μ = general mean, T_i = treatment's effects, i = control, T1 and T2, E_{ij} = the errors related to individual observation, M_j = the effect of time sampling post feeding j = zero and four hours, $(TM)_j$ = interactions between time and treatment.

Table 1. Ingredients (%) and chemical composition of concentrate feed mixture (CFM) and wheat straw.

| Ingredients composition % | Concentrate feed mixture (CFM) | Wheat straw |
|-------------------------------------|--------------------------------|-------------|
| Yellow corn | 39.9 | - |
| Wheat bran | 31 | - |
| Corticated cotton seed meal | 26 | - |
| Limestone | 2 | - |
| Salt | 1 | - |
| Vitamins and trace minerals mixture | 0.1 | - |
| Chemical composition % | | - |
| DM | 94.83 | 98.50 |
| OM | 88.88 | 86.34 |
| CP | 13.27 | 3.11 |
| CF | 19.22 | 27.16 |
| EE | 4.08 | 2.87 |
| NFE | 52.30 | 53.20 |
| Ash | 11.12 | 13.66 |

DM: Dry matter, OM: Organic Matter, CP: Crude protein, CF: Crude Fiber, EE: Ether Extract, NFE: Nitrogen free extract.

Results and Discussion

Nutrient digestibility and nutritive values

The effect of adding enzyme mixture to rams on the digestibility of nutrients and feeding values is presented in the Table (2). The digestion coefficients of DM, OM, CP, EE, CF, NFE and the feeding value expressed as TDN and DCP were not significantly affected by enzymes mixture supplementation. However, most nutrients digestibility tended to be numerically higher in treated groups than control. The findings of our research were approved by Ahmed *et al.* (2014) discovered that the nutrient digestibility and nutritive value were not affected with the supplement of different levels of phytase enzyme to sheep rations. Similarly, Wahyuni (2012) found that apparent digestibility of DM, OM and CP were not affected by supplement of enzymes to goats' diet. Moreover, the enzymes addition at rate 12g/d to sheep did not affect the DMI and feed digestibility as mentioned by Giraldo *et al.* (2009). López-Aguirre *et al.* (2016) found that supplemental exogenous enzymes to lambs' rations did not improve the digestibility of DM, OM, NDF, and CP.

Table 2. Effect of enzymes mixture supplement to rams on nutrient digestibility and feeding value (%)

| Items | Treatments | | | P-Value |
|-------|-------------|-------------|-------------|---------|
| | C | T1 | T2 | |
| DM | 53.47± 1.84 | 57.03± 2.72 | 57.16 ±7.66 | 0.832 |
| OM | 59.60± 1.79 | 62.65± 2.38 | 62.60± 6.72 | 0.848 |
| CP | 74.93± 1.14 | 77.67± 1.25 | 76.64± 4.43 | 0.783 |
| CF | 57.17± 3.52 | 58.12± 4.49 | 49.12± 5.96 | 0.399 |
| EE | 69.67± 3.08 | 78.23± 3.70 | 72.99± 1.40 | 0.190 |
| NFE | 53.47± 1.84 | 57.03± 2.72 | 57.16± 7.66 | 0.832 |
| TDN | 53.28± 1.91 | 56.23±2.30 | 54.04± 5.72 | 0.848 |
| DCP | 8.47±0.13 | 8.78±0.14 | 8.66± 0.50 | 0.783 |

C: Control rams received basal ration. T1: Rams received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Rams received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM). TDN: Total digestible nutrients, DCP: Digest Crud protein

Blood metabolites

The data of plasma components are summarized in Table (3). The results revealed that the mean values of total protein for ewes received enzymes mixture in the T1 group were ($P < 0.05$) higher than T2 and control groups. However, no significant differences were observed between treated groups. The ewes received high level of an enzymes combination (T2) were higher ($P < 0.05$) in albumin concentration and albumin / globulin ratio (A/G) compared to other groups. The globulin concentration was increased with addition of low level of enzymes mixture (T1) to ewes ration when compared with those received high level (T2) of enzymes mixture. The other blood metabolites like glucose, cholesterol, and triglycerides were not significantly affected by supplements enzyme mixture to ewes' rations. The value of total protein in the current study was considered in the normal range of sheep (6 -8 g/dl) as stated by Kaneko (1989). The higher total protein and albumin supplement enzyme mixture to ewes' rations may be due to higher protein intake, which increases amino acids available for absorption and metabolism (Baillet *et al.*, 1998). Also, Hallford *et al.* (1982) attributed the increases in serum albumin concentration due to higher protein intake. Similarly, Harper (1975) reported that the liver uses amino acids to create serum albumin. It could observe that the glucose values were higher in all groups of ewes, this may be attributed to the higher proportion of propionate, and propionate is the main glucose precursor in ruminants, which at the end of lactation the animals' requirement for glucose was lower (Ellis *et al.*, 2012; Polizel *et al.*, 2019). In the same context, Shetaewi and Daghsh (1993) found that the serum glucose level tended to be higher ($P < 0.09$) in lactating ewes (65.05 mg/dl) than in pregnant ewes (58.46). These results are in agreement with this reported by Marwan *et al.* (2019) and Ahmed *et al.* (2014), who sated an increase ($P \leq 0.05$) in total protein and albumin, with no differences in triglycerides, aspartate aminotransferase (AST), alanine aminotransferase (ALT) and creatinine for calves fed supplement of enzymes mixtures when compared with control. Also, the same effects were noted by Beigh *et al.* (2018) in lambs, El-Bordeny *et al.* (2015) and Peters *et al.* (2015) in dairy cows fed a ration that included

exogenous enzymes. However, Ahmed *et al.* (2014) stated that most blood metabolites were not affected by phytase addition among all groups.

Table 3. Effect of adding enzymes mixture to ewes on blood metabolites

| Items | Treatments | | | P- Value |
|------------------------------|---------------------------|---------------------------|--------------------------|----------|
| | C | T1 | T2 | |
| Total protein (g/dl) | 6.30 ^b ± 0.20 | 8.58 ^a ± 0.23 | 6.62 ^b ± 0.50 | 0.001 |
| Albumin (g/dl) | 3.24 ^b ± 0.28 | 3.55 ^{ab} ± 0.07 | 4.14 ^a ± 0.31 | 0.056 |
| Globulin (g/dl) | 3.06 ^{ab} ± 0.23 | 5.03 ^a ± 0.178 | 2.48 ^b ± 0.64 | 0.001 |
| A/G | 1.11 ^b ± 0.16 | 0.67 ^b ± 0.03 | 1.95 ^a ± 0.53 | 0.038 |
| Glucose (g/dl) | 173.15± 6.42 | 149.69± 4.77 | 148.74± 10.96 | 0.074 |
| Cholesterol (mg/dl) | 174.54± 4.14 | 162.53± 8.55 | 181.86± 3.29 | 0.091 |
| Triglycerides (mg/dl) | 173.61± 7.41 | 177.11± 10.77 | 174.73± 4.36 | 0.951 |

a,b, Significant differences between values in the same row with various superscripts ($P < 0.05$)

C: Control ewes received basal ration. T1: Ewes received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Ewes received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM).

Milk yield and milk components

The results of the enzymes mixture supplementation on milk yield of ewes were given in Table (4). The average milk yield was higher ($P < 0.05$) in the T2 group than control one. However, no differences were found between T1 and T2. Throughout lactation weeks, all treatment groups were higher in milk yield than the control, particularly in the fourth week of lactation. The increase in milk yield of ewes feed enzymes mixture may be a result of increased Dry matter intake Table (7) and ruminal fermentation (Khattab *et al.*, 2011) which led to increased energy available for milk production (Yang *et al.*, 1999) or due to higher production of TVFAs and NH₃ in the rumen, greater nutritional digestibility, and plasma glucose levels, which might result in the transfer of more glycolytic precursors to the udder (Azzaz *et al.*, 2017).

Table 4. Effect of enzymes mixture supplement on average milk yield (g) from the second week of birth to weaning

| Milk yield (g) | Treatments | | | P-Value |
|---------------------------|-----------------------------|-------------------------------|-----------------------------|---------|
| | C | T1 | T2 | |
| 2 wk. | 525.71± 47.64 | 577.86± 26.92 | 642.86± 46.95 | 0.166 |
| 4 wk. | 610.00 ^b ± 68.37 | 735.714 ^{ab} ± 44.99 | 830.88 ^a ± 51.36 | 0.039 |
| 6 wk. | 554.29 ^b ± 43.20 | 707.14 ^a ± 50.70 | 722.86 ^a ± 49.48 | 0.043 |
| 8 wk. | 447.857± 43.95 | 467.86± 48.59 | 539.29± 44.06 | 0.351 |
| 10 wk. | 403.571± 52.64 | 371.43± 35.42 | 467.14± 46.99 | 0.341 |
| 12 wk. | 295.714± 20.86 | 292.86± 25.91 | 356.43± 41.16 | 0.277 |
| Average milk yield | 472.86 ^b ± 34.76 | 525.48 ^{ab} ± 23.43 | 593.10 ^a ± 39.82 | 0.042 |

a,b Significant differences between values in the same row with various superscripts ($P < 0.05$)

C: Control ewes received basal ration. T1: Ewes received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Ewes received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM).

The results in Table (5) showed that the milk fat increased ($P < 0.05$) in the T1 than in the T2 groups. However, the differences were not significant between

T1 and control or between T2 and control groups. The other milk components were not affected by supplements enzymes mixture to ewes rations. The improvement in milk fat content of ewes fed enzymes supplement may be attributed to the larger amount of fiber intake, which could supply more acetate for fatty acid production, and or maybe as a result of the increase in energy and fatty acid availability for fat synthesis. (Mohamed *et al.*, 2013).

Table 5. Effect of enzymes mixture supplementation on milk composition

| Items (%) | Treatment | | | P-Value |
|------------------|---------------------------|--------------------------|--------------------------|---------|
| | C | T1 | T2 | |
| Milk Protein | 4.99 ± 0.26 | 5.070 ± 0.07 | 4.878 ± 0.18 | 0.710 |
| Milk Fat | 5.60 ^{ab} ± 0.44 | 6.23 ^a ± 0.23 | 5.06 ^b ± 0.18 | 0.044 |
| Milk Lactose | 4.72 ± 0.24 | 4.400 ± 0.24 | 4.620 ± 0.17 | 0.578 |
| Milk Total solid | 16.12 ± 0.65 | 16.388 ± 0.47 | 15.262 ± 0.34 | 0.291 |
| Milk Salt | 0.78 ± 0.04 | 0.700 ± 0.08 | 0.766 ± 0.03 | 0.550 |
| Milk S N F | 10.50 ± 0.54 | 10.155 ± 0.36 | 10.252 ± 0.38 | 0.839 |

a,b, Significant differences between values in the same row with various superscripts (P < 0.05)

C: Control ewes received basal ration. T1: Ewes received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Ewes received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM). SNF=Solids Not Fat.

The results of our study were proved by Titi and Lubbadah (2004), who reported that milk yield, total solids, fat, and protein increased (P<0.05) for sheep fed diet treated with fibrolytic enzyme. Similarly, Yang *et al.* (1999) found that the cows fed the high dosage of cellulase and xylanase enzymes were (P<0.05) higher for milk production than cows fed the control diet, with little effect on milk components. Moreover, the addition of commercial fibrolytic enzyme products to the diets of buffaloes increased milk production and milk fat (Kholif and Aziz., 2014). Mohamed *et al.* (2013) showed that supplementation of exogenous fibrolytic enzymes to dairy cows' rations increased significantly (p<0.003) milk production compared to the control group, without a significant change in the intake of dry matter. The cows fed enzyme total mixed ration tended (P<0.09) to be higher in milk fat and protein than cows fed the control diet (Dean *et al.*, 2013).

Rumen fermentation

Results of ruminal pH, TVFA and NH₃ -N concentration are presented in Table (6). The data showed that the pH value in the rumen fluid of the treated groups that received enzymes mixture decreased (P<0.05) as compared with the control one. The decreased ruminal pH in treated groups may be related to an increase in TVFA's production in these groups compared with the control group. ElKholany (2004) reported that the main reason for decreasing rumen PH is mainly due to the increase in TVFA's production in the rumen, this is further compounded by the rapid microbial breakdown of soluble carbohydrates (Balch,1977). However, no significant difference was observed between the T1 and T2 groups. Regarding the effect of sampling time on the rumen pH value, it was decreased (P <0.05) at 4 hrs after feeding than before feeding. These outcomes agree with those indicated by Azzaz *et al.* (2019), who found that the

pH value was lower after feeding with supplementation enzymes to goats' diet. The decrease in ruminal PH post-feeding could be related to the increase in the TVFA concentration as presented in Table (6). Huard *et al.* (1998) revealed that total VFA production accounted for 36% of the variance in rumen pH, which was inversely correlated with total VFA. Also, Owens *et al.* (1998) found that the extensive fermentation process of both nonstructural and structural carbohydrates and the production of VFA affect the rumen pH.

Table 6. Effect of enzymes mixture supplementation on rumen parameters

| Item | Treatments | Hours after feeding | | Means of treatment | P-value |
|------------------------|------------|---------------------------|----------------------------|--------------------------|--------------|
| | | Before feeding | 4 hrs. after feeding | | |
| pH | C | 7.21± 0.09 | 7.07 ^X ± 0.13 | 7.14 ^a ±0.07 | 0.003 |
| | T1 | 7.15 ± 0.02 | 6.70 ^Y ± 0.03 | 6.92 ^b ±0.07 | |
| | T2 | 7.31± 0.063 | 6.68 ^Y ± 0.06 | 6.99 ^b ±0.10 | |
| Means of time | | 7.22 ^A ±0.04 | 6.82 ^B ±0.06 | | |
| P- value | | 0001 | | | |
| TVFA's meq/100 ml/R. L | C | 9.25 ^Y ± 0.112 | 10.50 ^Y ± 0.129 | 9.88 ^b ±0.21 | 0.004 |
| | T1 | 9.75 ^X ± 0.111 | 11.00 ^X ± 0.18 | 10.38 ^a ±0.21 | |
| | T2 | 8.92 ^Y ± 0.24 | 10.83 ^{XY} ± 0.25 | 9.88 ^b ±0.33 | |
| Means of time | | 9.31 ^B ±0.12 | 10.78 ^A ±0.12 | | |
| P- value | | 0001 | | | |
| NH3 ml /100ml R. L | C | 12.07 ^Y ±0.33 | 12.02 ^Y ±0.13 | 12.04 ^b ±0.17 | 0001 |
| | T1 | 15.02 ^X ±0.16 | 14.68 ^X ±0.16 | 14.85 ^a ±0.12 | |
| | T2 | 14.18 ^X ±0.43 | 14.30 ^X ±0.49 | 14.24 ^a ±0.31 | |
| Means of time | | 13.76± 0.35 | 13.67± 0.33 | | |
| P- value | | 0.7340 | | | |

Means in row or Colum with the same letter (A, B), (a,b) and (x,y) were significantly at level of (0.05). C: Control ewes received basal ration. T1: Ewes received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Ewes received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM).

The ruminal TVFA s was higher ($P < 0.05$) in ewes that received the low level of enzymes mixture (T1) diet than other groups. However, no significant deference was found between T2 and control groups. As regarding the impact of sampling time on total VFA concentrations, the concentrations of TVFAs were decreased before feeding and increased post-feeding. The higher concentration of total VFA s reflected the higher fermentation activity in the rumen (Abdullah *et al.*, 1995). Also, the higher rumen TVFA s with enzyme addition in our study has resulted in higher availability of fermentable soluble carbohydrates. This result was approved by Pinos-Rodríguez *et al.* (2002), who showed that the addition of 5 g fibrolytic enzymes increased TVFA s concentration in the rumen of lambs with intra-ruminal. Similarly, Gado *et al.* (2009), Salem *et al.* (2013) and Bhasker *et al.* (2013) stated that the animals fed a diet with fibrolytic enzymes supplements were higher in total and individual VFAs concentrations.

The ruminal NH₃ -N concentration was increased ($P < 0.05$) in the rumen for ewes receiving enzymes treated groups than the control. The impact of the sampling time on NH₃ -N indicated that the values of NH₃ -N were not affected

between before and feeding after. The treated groups of ewes fed enzymes mixture that has protease enzymes and/or there was a change in the microbial community, supports its capacity to enhance rumen protein breakdown and greater NH_3 -N were released.

The results were similar to those reported by Bhasker *et al.* (2013) and Salem *et al.* (2013), who reported that the addition of exogenous enzyme at 40 g/head/d to stress increased ($p < 0.05$) concentration of rumen NH_3 -N and TVFA s before and 3h after feeding. Also, Azzaz *et al.* (2019) showed that xylanase and phytase supplementation increased the in vitro dry matter and organic matter degradability and ruminal NH_3 and TVFA s. However, Tseu *et al.* (2022) stated that the inclusion of different exogenous enzymes or their combination with ruminant diets did not affect nutrient digestibility, ruminal pH, ammonia nitrogen, and methane production.

Table 7. Effect of supplemental enzymes mixture on body weight of lambs and ewes feed intake

| Items | Treatment | | | P-value |
|--------------------------------|----------------------------|---------------------------|----------------------------|---------|
| | C | T1 | T2 | |
| Birth weight, Kg | 4.08± 0.27 | 4.16±0.17 | 4.09±0.17 | 0.955 |
| 15 d | 6.77± 0.44 | 6.58±0.24 | 6.62±0.32 | 0.921 |
| 30 d | 8.72± 0.62 | 9.18±0.32 | 8.67±0.42 | 0.707 |
| 45 d | 10.60± 0.72 | 11.49±0.34 | 10.30±0.63 | 0.349 |
| 60 d | 12.21± 0.90 | 13.91±0.35 | 13.27±0.61 | 0.210 |
| 75 d | 13.15± 0.97 | 15.26±0.45 | 15.04±0.49 | 0.080 |
| 90 d | 14.52 ^b ±1.15 | 17.25 ^a ±0.37 | 16.35 ^{ab} ±0.38 | 0.049 |
| Daily gain, g/d | 116.10 ^b ±10.41 | 145.44 ^a ±3.21 | 136.14 ^{ab} ±4.91 | 0.022 |
| Total gain, kg | 10.44 ^b ±0.94 | 13.09 ^a ±0.29 | 12.25 ^{ab} ± 0.44 | 0.022 |
| Dry matter intake, kg/d | | | | |
| Concentrate | 0.753± 0.03 | 0.754±0.03 | 0.754±0.02 | 0.999 |
| Roughage | 0.33 ^b ± 0.01 | 0.43 ^a ± 0.01 | 0.46 ^a ± 0.02 | 0.001 |
| TDMI | 1.08± 0.04 | 1.18±0.04 | 1.21± 0.04 | 0.094 |

^{a,b}, Significant differences between values in the same row with various superscripts ($P < 0.05$)

C: Control ewes received basal ration. T1: Ewes received low level of enzymes supplement (30 g enzyme mixture /100 kg CFM), T2: Ewes received high level of enzymes supplement (60 g enzyme mixture /100 kg CFM). TDMI= Total Dry Matter Intake.

Lambs' performance

The average body weight of newborn lambs of experimental ewes did not significantly affect during the nursing stage (Table 7). However, the body weight and average growth rate of lambs were higher ($P < 0.05$) in T1 than in the control group, with no difference, were found between T1 and T2 or T2 and control groups. The improved daily gain of lambs with enzyme supplements to ewes might be related to the increased milk production and milk fat in treated groups. The current results were confirmed with those observed by Gado *et al.* (2011) showed that treating ensiled orange pulp with the same substance in the present research enhanced lambs' live weight growth, nutritional digestibility, and ruminal fermentation. Also, Titi and Lobbied (2004) found that lambs' birth

weights were not significantly impacted by fibrolytic enzyme treatments to ewes, however, the weaning weights of lambs have increased ($P < 0.05$) of treated dairy goats and ewes.

The results in Table (7) demonstrated that the enzymes mixture supplement to ewes' rations did not reveal any significant effect on total DM intake during the experimental period. However, the roughage intake was increased ($P < 0.05$) with groups that received enzymes mixtures when compared with the control group. The increased roughage intake in the current study may be due to increased microbial populations and colonization of the rumen content after enzymes treatment (Beauchemin *et al.*, 2000).

Conclusion

Exogenous enzymes mixture supplement to ewes' diet during the last two months of pregnancy and lactating period, particularly at a rate of 30 g/100 kg concentrate mixture improved milk yield, milk fat, some blood metabolites, rumen fermentation, and the weight of newborn lambs at weaning time. However, these supplements were less effective on nutrient digestibility and feeding value of rations. Therefore, our study could be recommended to add an enzyme mixture to the ewes' diet at the rate of 30 g/100 kg concentrate mixture during the last two months of pregnancy and during the weaning period of lambs.

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تأثير إضافة مخلوط الأنزيمات على الأداء الإنتاجي للنعاج وحملاتها

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ملخص

تتحرى الدراسة تأثير إضافة مخلوط الأنزيمات للنعاج والكباش المحلية على كفاءة هضم الغذاء ومقاييس سائل الكرش وإنتاج اللبن وتركيبه وبعض مقاييس الدم بالإضافة الي أداء الحملان. استخدمت في هذه الدراسة 21 نعجة عشار وقبل شهرين من الولادة المتوقعة وزعت بصوره عشوائية الي 3 مجاميع متساوية طبقا لمتوسط وزن الجسم بكل مجموعة 7 نعاج. مجموعة المقارنة (الكنترول) غذيت بمخلوط العلف المركز (عليقة اساسية) بدون إضافة مخلوط الانزيمات، المجموعة الثانية (T₁) والثالثة (T₂) غذيت بالعليقة الأساسية بالإضافة الي 30 و60 جرام من مخلوط أنزيمات لكل 100كجم علف مركز مخلوط على التوالي، واستمرت لمدة 3 شهور بعد الولادة. أظهرت النتائج ان إضافة مخلوط الأنزيمات لم يؤثر على معاملات هضم عناصر الغذاء والقيم الغذائية للعلائق، متوسط إنتاج اللبن خلال فترة الرضاعة ارتفع معنوي في المجموعة الثالثة (T₂) مقارنة بالمجموعات الأخرى ومع ذلك لم تكن هناك فروق معنوية بين (T₂) و(T₁)، او بين (T₁) والكنترول. النسبة المئوية لدهن اللبن زاد زيادة معنوية مع إضافة المستوي المنخفض بمخلوط الانزيمات مقارنة بالمستوي العالي. الاحماض الدهنية الطيارة الكلية في سائل الكرش ارتفعت في المجموعة (T₁) مقارنة بالمجاميع الأخرى، تركيز نيتروجين الامونيا للمجاميع المعاملة زاد معنوياً مقارنة بمجموعة المقارنة. البروتين الكلي والجلوبيولين في بلازما الدم زاد زيادة معنوية في المجموعة الثانية (T₁) مقارنة بالمجموعة الثالثة (T₂) ومجموعة المقارنة (الكنترول). متوسط الوزن اليومي للحملان زاد معنوياً في المجموعة الثالثة (T₂) مقارنة بمجموعة الكنترول. تخلص نتائج هذه الدراسة، أنه يمكن إضافة مخلوط الأنزيمات لعلائق النعاج خلال الشهرين الأخيرين من الحمل بمعدل 30جم /100كجم علف مركز فهذا يحسن من إنتاج اللبن ودهن اللبن وبعض مكونات الدم وتخمرات الكرش ووزن الحملان حديثي الولادة وعند الفطام.

الكلمات المفتاحية: مخلوط الانزيمات، هضم العناصر الغذائية، انتاج اللبن، تخمرات الكرش، اداء الحملان.