UTILIZATION OF ALLZYME SSF TO IMPROVE THE NUTRITIVE VALUE OF OLIVE CAKE IN SHEEP

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SUMMARY

The objective of the current work was to study the effect of adding different level of allzyme SSF to olive cake based diet of sheep on the intake, digestibility and nutritive value. Twenty four Barki rams (average 40.0 ± 2.11 kg live body weight) at 2-3 years old were used in the experiment. Animals were randomly allocated to four groups (6 animal/group) in separate pens and offered the experimental diets . All animals were fed concentrate feed mixture (CFM) consists of 90 % olive cake, 7% molasses, 1% sodium chloride, 1.5 % Limestone, 0.5 % mineral. During this trial the experimental diets were offered at (85% CFM and 15% Barseem hay) .in which olive cake were treated with 0.5, 1.0 and 1.5% of the allzyme SSF in the diet, respectively, for D2, D3, and D4 groups in comparison with the control (D1, without enzyme addition). The results indicated that treating olive cake with allzyme SSF decreased its fiber content. Animals in D3 and D4 groups contained relatively similar content of dry matter (DM), crude protein (CP), fiber fractions and condensed tannins. The results indicated that palatability of olive cake was improved with allzyme SSF addition. The apparent digestibility of all nutrients were higher in D3 and D4 than that in D2 and the control group (D1). The highest TDN and DCP intakes were reported for sheep in D3 and D4 followed by D2 and then D1. Results showed that TDN and DCP values were increased with increasing the level of enzyme in the diets. The concentration of ruminal pH was decreased , while that of ammonia and volatile fatty acids were increased gradually from zero to 4 hours post feeding. No significant differences were found in serum total protein, albumin and globulin concentrations among sheep in the four treated groups. Blood concentrations of creatinine, glutamic pyruvic transaminase (GPT) and glutamic oxalacetic (GOT) were not affected significantly by Allzyme SSF addition. It could be concluded that using Allyzme SSF supplementation at levels 1.0 and 1.5% in sheep diets tended to improve the digestibility and the nutritive values of sheep diet.

Keywords*: olive cake, digestibility, nutritive value, allyzme SSF, blood metabolities*

INTRODUCTION

The feed cost represents more than 70% of the total production cost. It is now urgent to look for alternative feed stuff to compensate the high cost of the conventional feedstuff. The gap between the availability and requirements of animal feed in Egypt is about 9 million tons of dry matter equivalents to almost 4 million tons of total digestible nutrients (TDN) per year (Bendary *et al.,* 2006), therefore efforts allowed some by products and organic wastes with the aim of decreasing the animals feed shortage. In Egypt, there are about 119,000 Fadden planted with olive trees, produce about 314,450 tons of olive (Ministry of Agriculture (2012) and land reclamation, basically, one ton of olive produce approximately 350 kg of crude olive cake. Olive cake is usually described as low quality feedstuff because of their low nutritive value, high content of fiber (Molina-Alcaide and Nefzaoui,1996; Abbeddou et al.,2011) and condensed tannins (Martin Garcia *et al*., 2003), low degradability of cell wall component (Felya *et al*., 2006; Teimouri Yansari *et al*., 2007) and low content of protein and energy (Al-masri and Guenther, 1995; Molina Alcaide and Nefzasuir, 1996 and abbeddau *et al*., 2011). Different approaches to improve the nutritive value of olive cake have been used. De. Stoning (manual removal of crushed needs) ensiling (Hadjipanayiotou ,1994; Aljassim *et al*.,1997; Abo omar *et al*., 2012), pelleting (Abo omar *et al* 2012), and exogenous fibrolytic enzymes (Awawdeh and Obeidat, 2011).; Bedford and Partridge 2011 found that supplementing the feed with specific enzymes improves the nutritional value of feed ingredients,

increasing the efficiency of digestion. Also, feed enzymes help break down anti –nutritional factors that presents in many feed ingredients and reducing cost

The objectives of the current work were to study the effect of different level of allzyme SSF in the olive cake based diet of sheep on the chemical composition, intake, digestibility and nutritive value.

MATERIAL AND METHODS

The present study was carried out at Ras Sudr experimental research station, Desert Research Center. Fresh olive cake was collected from modern olive oil extraction at Ras Sudr area. Olive cake is a residue detained at the mill after extracting the oil by pressing the whole olive fruits, thereafter, crude olive cake was dried simply by sun drying till about 10% moisture content was attained. Then, olive cake was screened (2mm screen) after drying and stored in bags to be used in making dietary treatments.

The current experiment was based on treating the olive cake with different levels of allzyme SSF in the diet. The allzyme SSF enzyme is consisted of Phytase Min 300 SPU/g, Protease Min 700 Hut/g, Cellulase Min 40 SMcU/g, Xylanase Min 100 Xu/g, Betaglucanase Min 200 PGU/g, Amylase Min 30 FAU/g and Pectinase Min 4000 AJDU/g .olive cake was treated with mixed to seven days under air temperature in shaded place then dried also under air temperature for one week in sunny place then fed to animals direct .Twenty four Barki rams (average 40.0 ± 2.11 Kg live body weight) at 2-3 years old were used in the experiment. Animals were randomly allocated to four groups (6 animal/ group) in separated pens and offered the experimental diets. All animals were fed concentrate feed mixture (CFM) supplement was formulated with 90 % olive cake, 7% molasses, 1% sodium chloride, 1.5 % Limestone, 0.5 % mineral. During this trial the experimental diets were offered at (85% CFM and 15% Barseem hay) for 45 days as a feeding trial

Each group was offered one of the four diets as olive cake without enzyme (control group, D1), while the second, third and fourth groups were fed olive cake supplemented with 0.5 ,1.0 and 1.5% Allzyme SSF, respectively. The Allzyme SSF was supplemented to the concentrate supplement by spraying with 0.5kg, 1.0kg and 1.5 kg/ ton of allzyme SSF solution per ton of the concentrate supplement with D2,D3 and D4 respectively.

The concentrate supplement and barseem hay were offered in separate feeders at 08:00 h daily to cover almost 110% of the maintenance requirements as recommended by Kearl (1982) for sheep in developing countries. Voluntary feed intake was recorded daily for each group. Amounts of feed offered and refusal were recorded to estimate the actual voluntary feed intake for each group. All animals were weighted at the beginning and end of the feeding trial. Drinking water was available for sheep all time.

At the end of the feeding period, the digestibility trial were conducted using four animals per group those were placed individually in metabolism cages for 14 days. The first 7 days were preliminary period, while the last 7 days were for the collection period. The average voluntary intake from olive cakes during the last week of the feeding period was offered to animal during the digestibility trial. During the collection period, voluntary intake of each animal was recorded daily. Total fecal output was collected and sampled daily (almost 10% of the total). Samples of feed, orts and feces were dried as 65°C for the dry matter (DM) content. Feces samples were composited separately for each animal and stored for chemical analysis.

Analytical methods:

Proximate analysis of diets and feces were conducted according to the official procedure of A.O.A.C., (1990). The crude protein (CP) in offer, orts and feces was determined by micro Kjeldhl method of Chibnall *et al*., (1943). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined (Van Soest et al .,(1991). Tannin were analyzed according to Makkar and Googchild (1996). Rumen liquor was taken from each animal in the last day of the digestibility t rial for 2 consequent times (0 and 4 hours post morning feeding by stomach tube). The filtrate a as used in determination of pH immediately after the sampling using portable pH meter. Rumen liquor was used to determined ammonia nitrogen concentrations according to Abou-Akkada and EL-Shazly (1964), while total volatile fatty acids (TVFA's) concentrations was determined according to Warner (1964).

Data obtained in this study was statistically analyzed by a completely randomized design (SAS, 2004) Duncan multiple range test was also used to test the significant differences among treatments wherever possible (Duncan, 1955).

RESULTS AND DISCUSSION

Chemical composition:

The Effects of allzyme SSF addition on olive cake based concentrate supplement and its chemical composition are presented in Table (1). The supplemented olive cake with allzyme SSF reported a higher content of CP and a lower content of call wall component compared to that without allzyem SSF. Results are in agreement with Hadjipanayiotou (1999a, b). Both D3 and D4 diets contained relatively similar content of DM, CP, NDF, ADF, ADL and condensed tannins as reported by Awawdeh and Obeidat (2013) and Gomaa *et al.* (2016). The allzyme SSF addition decreased the olive cake NDF contents that probably due to breaking off gross linkage between lignin and cell wall component and solubilizing of cell wall contents (mainly hemicelluloses).

Table (1) Chemical composition of the exprimentale diets (% on dry matter basis)

Item	D1	D2	D3	D4	Hay
Dry matter	93.7	95.3	96.1	96.3	95.3
Crud protein	6.87	7.72	8.90	9.00	12.2
Ether extract	8.36	8.78	8.39	8.75	1.42
Crud fiber	22.5	17.2	16.3	14.1	30.7
N- Free extract	49.0	53.6	54.0	55.9	47.0
Ash	13.3	12.7	12.4	12.2	8.65
Cell wall components					
NDF	62.5	59.9	57.8	57.6	59.0
ADF	48.9	47.5	46.7	45.9	44.1
ADL	36.6	30.01	24.9	22.5	24.1
C.T	0.24	0.20	0.16	0.16	1.97

D1: Olive cake Untreated, D2: Olive cake + 0.5 % Allzyme SSF, D3: Olive cake + 1.0 % Allzyme SSF, D4: Olive Cake + 1.5 % Allzyme SSF, NDF: Neutral detergent fiber NDF, ADF: Acid detergent fiber, ADL: Acid detergent ligninnsed, C.T: Conde Tannins

Olive cake has high cell well constituents (NDF) *and* lingo – cellulose (ADF) and lignin (ADL) contents. It is known that NDF is a good indicator for diet intake. The low NDF value for olive cake as affected by allyzme SSF addition (Table 1) was reported by several investigators (Van soest, 1975, Hadjipanayiotou, 1999 a, b; cabiddu et al, (2004) and Lizhi Wang and Bai Xue (2016).

Feed intake:

Average daily feed intake values for the feeding trial are presented in Table (2). During the feeding trail, constant voluntary DM intake of olive cake with or without enzyme was achieved at same time after 21 days of feeding. The voluntary intake of sheep fed D2, D3 and D4 diets (Table 2) was increased gradually, then became stable at the third week of feeding up to the end of trail, indicating that all diets had the same acceptability. Generally, the results indicated that palatability of olive cake with allzyme SSF was higher than that without allzyme SSF. When the dry matter intake was expressed as a percent of body weight, sheep fed D2, D3 and D4 diets recorded higher values than those fed the control (D1) without allzyme SSF (2.56, 2.54, 2.55 vs. 2.10% respectively). Nefzaoui (1983) reported that olive cake is not very palatable and not widely consumed. Most of the tests recommended the inclusion of 8 to 10 percent of molasses with the olive cake. On the other hand, Awawdeh and Obeidat (2013) reported no effects of dietary treatments on lambs intake when they fed a diet containing olive cake. Findings are in agreement with the results of Bedford and Partridge, (2011).

Digestibility coefficients and feeding values:

The results of digestion coefficients and nutritive value of the experimental diets are presented in Table (3). Results showed that the inclusion of allzyme SSF in the diets significantly ($P<0.01$) improved the digestibility in comparison with the control diet (D1). Sheep fed D3 and D4 diets tended to digest better with more efficiently than those fed the control diet and D2 with low level of Allzyme SSF (0.5%). The reduction in the digestion of olive cake without allyzme SSF could be attributed to the high level of

fiber fraction content (Table 1). These observations are agree with those of reported by Soliman *et al*. (2007) and Manylouis and El-Banna (2013). The apparent digestibility of DM, CP and fiber fractions were higher for animals fed D3 and D4 than those fed D2 and control group (D1). The digestibility was significantly varied among feeding treatments, being higher for animals fed D2, D3 and D4 vs. those feeding the control group (D1)., particularly in the NDF digestibility that were higher for animals fed D3 than those fed D1, D2 and D4, while the ADF and ADL were lower for D4 versus D3, D2 and D1 that may be due to the increase of Allzyme SSf in D4 . This may be supported by O'Connor et al.,(2007) who found that the digestibility was decreased when cellulase was added to high quality forage diets of horses.

Table (2) Voluntary intake of experimental diet of sheep

Item	D1	D2	D3	D4	±SE
Voluntary intake, g /Kg BW					
Dry matte intake					
Supplement g/Kg BW	17.8^{b}	22.2^{a}	22.0^{a}	22.1^{a}	0.06
Hay g\kg BW	3.31^{a}	3.44^{a}	3.36^{a}	3.35^{a}	0.04
Total g\kg BW	21.1^{b}	25.6^{a}	25.4^{a}	25.5^{a}	0.09
Crud protein intake					
Supplement g/kg BW	1.22^{d}	1.71^{c}	1.77^{b}	1.77^{a}	0.004
Hay g/kg BW	0.40^{a}	0.42a	0.41^{a}	0.41^{a}	0.006
Total g/kg BW	1.63^{b}	2.13a	2.18^{a}	2.18^{a}	0.06
Ether extract intake					
Supplement g\kg BW	1.13^{d}	1.28^{c}	1.41^{b}	1.49^{a}	0.004
Hay g \kg BW	0.050^{a}	0.05^{a}	0.05^{a}	0.05^{a}	0.001
Total g\kg BW	1.18^{b}	1.33^{c}	1.47^{b}	1.55^{a}	0.004
Crud fiber intake					
Supplement g\ kg BW	4.01^{a}	3.82^{b}	3.60^{c}	3.12^{d}	0.14
Hay g\kg BW	1.01^{a}	1.05^{a}	1.03^{a}	1.03^{a}	0.01
Total g\kg BW	5.03^{a}	4.88^{b}	4.63^{c}	4.15^{d}	0.02
NDF: Neutral detergent fiber NDF					
Supplement g\ kg BW	11.13^{d}	12.87^{b}	13.23^{a}	12.31^{c}	0.04
Hay g\kg BW	1.96^{a}	2.03^{a}	1.98^{a}	1.98^{a}	0.03
Total g\kg BW	13.09^{d}	14.90b	15.21^{a}	14.29^{c}	0.05
ADF: Acid detergent fiber					
Supplement g\ kg BW	8.71^{d}	10.6^{a}	10.3^{b}	10.16^{c}	0.03
Hay g\kg BW	1.47a	1.51^{a}	1.48^{a}	1.47^{a}	0.02
Total g\kg BW	10.16^{d}	12.07^{a}	11.78^{b}	11.62^{c}	0.04

A, b, c and d: means within the same row with different superscripts are differ significantly (P<0.05)

Allzyme SSF is believed to be effective in improving nutrient digestibility of ruminants especially that of fiber. Ballard *et al*. (2003) reported that dietary addition of compound cellulose increased the apparent digestibility of DM, OM and NDF in dairy cows. It is known that nutrient digestibilities of Olive cake are low and variable (Molina-Alcaide and Yanez-Ruiz, 2008). It has been reported that DM, fiber, and CP digestibilities were low and fat digestibility was high (Molina –Alcaide and Nefzaoui,1996).

Bedford and Partridge(2011) showed that additive enzymes improved the digestibility of energy, protein and minerals in the feed. Also Hadjipanayiotou (1994) found that treating olive cake with urea improved its nutrients digestibility compared to crude olive cake, but the ensiling of urea treated olive cake did not further increase nutrient digestibility.

Results in Table (3) indicated that nutritive values of TDN and DCP were varied significantly (P<0.01) among the treated groups those fed olive cake with enzyme versus the control group. Allzyme SSF improved nutritive value of TDN by 23, 30 and 39 unites in sheep fed D2, D3 and D4 diets, respectively, compared to the control diet (D1).

Both TDN and DCP intakes were significantly (P<0.01) improved by Allzyme SSF addition. The highest TDN and DCP intakes were recorded for sheep in D3 and D4 followed by those in D2 and then D1, these may be due to the better digestion and utilization of diet with animals fed olive cake with enzyme. animals fed olive cake with allzyme SSF (D2, D3 and D4) were able to cover their maintenance requirement of energy (Kearl, 1982). Moreover, the TDN intake was exceeded by 12, 28 and 29% for D2,

D3 and D4, respectively and less by 11% for the control compared to the recommendation of Kearl (1982) for sheep in developing countries.

Table (3): digestion coefficients and nutritive value of the experimental diet of sheep

Item	D1	D2	D3	D4	±SE
Digestion coefficients %					
DM	43.8^{c}	52.3^{b}	57.6^{a}	55.5^{ab}	1.25
OM	42.6^{c}	51.8^{b}	53.4^{ab}	56.0^{a}	0.39
CP	25.9^{c}	39.4^{b}	43.8^{a}	45.8^{a}	1.06
CF	35.9^{c}	41.4^{b}	49.6^{a}	41.4^{a}	1.01
NFE	53.9^{b}	53.9^{b}	62.5^{a}	59.9^{a}	1.32
NDF	44.2^{b}	44.5^{b}	52.3^{a}	45.5^{a}	1.49
ADF	35.0^{b}	38.3^{b}	46.3^{a}	37.3^{b}	1.91
ADL	27.0^{a}	28.3^{a}	29.4^{a}	25.6^{a}	1.21
Total digestible nutrients					
TDN g\kg BW	9.99^{c}	12.6^{b}	14.5^{a}	14.6^{a}	0.28
% of DMI	47.3^{b}	49.0^{b}	56.9^{a}	57.3^{a}	1.17
% of MR*	89.0	112	128	129	-----
Digestible crude protein					
DCP g/kg BW	0.42^{d}	0.84^{c}	0.94^{b}	1.04^{a}	0.03
% of DMI	1.99^{c}	3.28^{b}	3.70^{a}	4.10^{a}	0.13
% of MR*	46.6	93.3	104	115	------

A, b, c and d: means within the same row with different superscripts are differ significantly (P<0.05)
MR maintenance requirements (Kearl, 1982)*

The nutritive values (TDN and DCP) of the treated diets were improved with addition of allzyme SSF. Results showed that TDN and DCP values were increased with increasing the level of enzyme in the diets. The TDN was improved by 49, 57 and 57% with addition of enzymes in the diets at 0.5, 1.0 and 1.5%, respectively, compared to the control diet. The same trend was reported by Beauchemin *et al*. (2003), Gado *et al*. (2007a), EL-Manylawi and EL-Banna (2013) and Lizhi Wang and Bai Xue (2016). The DCP was high with animal fed D4 diet followed by D3, D2, and then D1, due to the increase in the digestibility coefficients of CP in D4.

Rumen liquor parameters:

As given in Table (4) analysis of variance revealed that a significant (P<0.05) differences were found among the treated diets in pH at 4 hours post feeding in comparison with those recorded at zero time. Prasad *et al*., (1972) found that rumen pH is one of the most important factors affecting fermentation in rumen and influences its functions. In all diets, concentration of ruminal pH decreased gradually after 4 hours of feeding. Findings also have been reported in buffalo calves (Sabbah-Allam *et al*., 2013). Molina-Alcaide and Nefaoui (1996) indicated that feeding Olive caketo sheep resulted in favorable pH of fibrolytic activity (pH from 6.6 to 7.2).

Results of rumen ammonia and volatile fatty acids concentrations are presented in Table (4). In all diets, rumen ammonia concentration increased at 4 hours post feeding. These results may be due to the improvement of rumen fermentation. Wanapat *et al*. (1994) reported that ruminal ammonia nitrogen is an important nutrient in supporting the efficient of rumen fermentation and microbial protein synthesis. At the same trend of rumen ammonia, the level of ruminal total volatile fatty acids was increased gradually from zero time to 4 hours post feeding. This is in agreement with those obtained by ZaZa *et al*. (2013) and Sabbah-Allam *et al*. (2013).

Blood constituents:

Blood constituents as affected by treated diets are given in Table (5). No significant differences were found in serum total protein, albumin and globulin concentrations among sheep fed on the four treated diets. However, all values were within the normal range as obtained by Zaza *et al*. (2013) and Kaneko *et al*. (1997) who found that the normal level of plasma albumin concentration were 3 to 5 g/dl. The enzymatic activities of liver function, blood concentrations of GPT and GOT were not affected significantly by Allzyme SSF addition. Blood plasma enzymes activity (GPT and GOT) are the most important indicator of liver cell activity. Boots *et al*. (1969) reported that several factors may affect the two enzymes activities (GPT and GOT), such as the feeding practices, environment, genetic control,

response to stress, age, liver function and body weight. Creatinine was not significantly affected by dietary treatments. Urea-N (mg/dl) was significantly higher (P<0.05) with animal fed diets D1 and D2 compared with those fed D3 and D4 diets. Bayat *et al*. (2003) fed whole whey to 3-6 month old Holstein steers and found that plasma urea nitrogen levels were significantly lower with whey diets than the control diet (14.9 and 18,6 versus 25.2 mg/dl). Creatinine and urea concentration in blood are parameters used to asses the function of kidneys in human and animals.

Table (4): concentrations of rumen ammonia nitrogen and total volatile fatty acids of the experimental diet of sheep

Item	D1	D2	D3	D4	±SE
Rumen ammonia nitrogen mg/100 ml					
Before Feeding	10.89^{b}	10.73^{b}	11.92^{a}	11.34^{ab}	0.27
After 4 hours	11.22^{b}	12.28^{a}	12.77^{a}	11.75^{ab}	0.30
Change	0.33	1.55	0.85	0.41	-----
Total volatile fatty acids meq./100ml					
Before Feeding	4.36^{a}	3.92^{b}	3.97^{b}	3.66^{b}	0.28
After 4 hours	5.02^{a}	4.93^{a}	4.96^{a}	5.09^{a}	0.34
Change	0.66	1.01	0.99	1.43	----
PH					
Before Feeding	7.46^{a}	7.45^{a}	7.53^{a}	7.22^{a}	0.18
After 4 hours	6.95^{a}	6.70^{b}	6.91^{a}	6.87^{ab}	0.06
Change	0.51	0.75	0.63	0.35	------

A, b, c and d: means within the same row with different superscripts are differ significantly (P<0.05)

Table (5) Concentration of blood metabolites of sheep fed the experimental diet of sheep

Item	D1	D2	D3	D4	±SE
Urea nitrogen mg/100l	20.3^{ab}	21.7^{a}	16.9^{b}	17.4^{ab}	1.10
Creatinine mg\ 100l	0.734	0.726	0.707	0.839	0.05
GPT , U/L	77.8	74.5	85.6	73.2	6.18
GOT, U/L	208.0	195.0	205.0	193.0	14.4
Total protein g/dl	5.98	5.99	5.87	5.32	0.23
Albumin, g/dl	3.50	3.41	3.72	3.55	0.22
Globulin, g/dl	2.49	2.59	2.15	1.77	0.34

A, b, c and d: means within the same row with different superscripts are differ significantly (P<0.05)

CONCLUSION

Olive cake is not characterized as high palatable feed, so that the addition of 8-10% molasses can result in a high intake level. Olive cake alone can cause weight losses in the animal as it is also poorly digested. It causes low ammonia and low volatile fatty acids production as a proof of its low nutritive value. Therefore, some additives can improve the nutritive value and digestibility of olive cake. Based on the present results, it could be concluded that using Allyzme SSF supplementation at level 1.0 and 1.5% in sheep diets tended to improve the digestion coefficients and nutritive values of the olive cake. Future research should concentrate on finding other methods to improve the nutritive value of olive cake.

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الاستفاده من الانزيم (SSF) فى تحسين القيمة الغذائية لتفلة الزيتون فى الاغنام

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أجريت هذه الدراسة فى محطة بحوث راس سدر بهدف دراسة تأثير مستويات مختلفة من الانزيم وتأثيره على المأكول والتركيب الكيماوى ومعاملات الهضم والقيمة الغذائية فى عليقة الاغنام .تم استخدام عدد24 كبش برقى متوسط اوزانهم 40 كجم ومتوسط اعمارهم من 2-3 سنوات وزعت عشوائيا الى 4 مجاميع فى كل مجموعة 6حيوانات وتم تكوين عدد4 علائق تتكون 85% تفلة زيتون و15% دريس برسيم على اساس المادة الجافة. المجموعة الاولى تعتبر المجموعة الضابطة لا تحتوى على انزيم اما المجموعة الثانية والثالثة والرابعة اضيف اليها أنزيم بمستوىSSF 0.5 % – 1 % – 1.5 %على التوالى.

أظهرت نتائج الدراسة ان تفلة الزيتون مع الانزيم خفضت الالياف الخام و NDF فى كل من المجموعتين الثالثة والرابعة. مكونات تفلة الزيتون من المادة الجافة والبروتين الخام ومكونات الالياف والتانينات كانت متشابهة . تفلة الزيتون مع الانزيم ذات استساغة عالية بالمقارنة بتفلة الزيتون بدون انزيم. معامل هضم كلا من المادة الجافة والبروتين الخام و ,NDF كانت عاليه فى المجموعة الثالثة والرابعة بالمقارنة بالمجموعة الثانية والاولى (الكنترول). المركبات الكلية المهضومة والبروتين المهضوم كانت النسبة عالية فى المجموعة الثالثة والرابعة عن المجموعة الثانية والاولى على التوالى وايضا كانت المركبات الكلية المهضومة والبروتين المهضوم يزيد بزيادة مستوى الانزيم فى العليقة .بالنسبة لمقايس الكريش نجد ان درجة الحموضة تقل بعد 4 ساعات من الاكل بينما الامونيا والاحماض الدهنية الطيارة فانها تزيد بعد 4 ساعات من الاكل بالنسبة لاختبارات مكونات الدم لاتوجد فروق معنوية فى البروتين الكلى والالبيومين والجلوبيولين وكذلك وظائف الكبد والكرياتينين.ويمكن ان نستخلص من هذه الدراسة بأن اضافة الانزيم بمستوى 1% و 1.5% فى عليقة الاغنام حسن من معاملات الهضم والقيمة الغذائية .