

## IMPACT OF PARTIAL REPLACEMENT OF COTTONSEED MEAL BY NIGELLA SATIVA MEAL ON NUTRIENTS DIGESTIBILITY, RUMEN FERMENTATION, BLOOD PARAMETERS, GROWTH PERFORMANCE OF GROWING LAMBS

M.A.M. Abdullah and M.M. Farghaly

Department of Animal Production, Faculty of Agriculture, Assiut University, Assiut, Egypt.

### SUMMARY

This study was conducted to evaluate the effect of partial replacement of cotton seed meal in concentrate mixture by *Nigella sativa* (*N. sativa*) meal on nutrients digestibility, rumen fermentation, blood metabolites and performance of lambs. Eighteen of healthy Farafra male lambs aged between 5 and 6 months old and had average of  $27.2 \pm 1.3$  kg body weights. The lambs were randomly assigned into three groups, 6 animals each lasted for 90 days. The experimental groups were a control group (CON) fed basal diet containing concentrate mixture plus wheat straw and treated groups were fed basal diet plus 33.3% (T1) and 66.7% (T2) as a substitution of cottonseed meal by *N. sativa* in concentrate mixture. The lambs were fed 65% of their requirements as a concentrate mixture based on NRC guidelines and the rest were covered from wheat straw which given as roughages ad libitum. Three digestible trials were conducted with twelve mature local rams, distributed into three group and using three experimental diets, CON, T1 and T2. In growth trial body weight were recorded and daily gain were calculated. Plasma metabolites, ALT, AST enzymes, thyroid hormones (T3 and T4) and total antioxidant capacity (TAC) were determined in blood samples. In digested trial rumen liquor samples were collected to measure the protozoal count, pH, ammonia N and total VFAs concentrations. Feed and fecal samples were analyzed using AOAC (2012). The results were statistically analyzed using of SPSS statistical package. The results refer to the body weight and daily gain in T2 group was increased as compared with T1 and CON groups. The total DM and CP intake were higher ( $P < 0.05$ ) in T2 group as compared with CON group. The partially substitution of cotton seed meal by *N. sativa* meal increased ( $P < 0.05$ ) the concentrations of NH<sub>3</sub>-N and total VFAs, while the total protozoal count in rumen fluids was decreased ( $P < 0.05$ ) as compared with CON group. Also, the plasma cholesterol and ALT enzyme was decreased ( $P < 0.05$ ) in *N. sativa* meal groups compared to control group. However, T3 hormone and total antioxidant capacity (TAC) were higher ( $P < 0.05$ ) in *N. sativa* groups than control group. The nutrient digestibility was increased ( $P < 0.05$ ) with increasing substitution rate from 33.3 to 66.7% of cotton seed meal by *N. sativa* meal. It can be concluded that the partial replacement of cottonseed meal in concentrate mixture by *N. sativa* can be used as an unconventional protein source in lambs' rations with no any harmful effect on performance of growing lambs..

**Keywords:** *Nigella sativa* meal, nutrients digestibility, rumen fermentation, blood metabolites, lambs, performance.

### INTRODUCTION

Currently, consumers demand the safety and the quality of feeding. So, the use of feeds containing bioactive compounds such as thymol, flavonoids, saponins, limonene, thymoquinone, essential oils and others are responsible for the healthy animal product. Several efforts of research in ruminant production to identify strategy able to improve the quality of livestock product. On the other hand, one of the hot points in the advanced research is the stability of animal product against the oxidative deterioration process that limits the shelf life (Faustman et al., 2010). Herbal medicinal plant concentrated by bioactive compound in different parts, such as leaves, roots and seeds which can be used for these purpose in animal feed.

One of the herbal plants is *N. sativa* or black cumin. Black seed is widely grown in the Middle East and Southeast Asia, Mediterranean area and recently Eastern Europe. Black seed belongs to family *Ranunculacea*, which contains volatile oils, alkaloids and a variety of phytochemical compounds. The chemical composition of *N. sativa* are crude protein 38.7% crude fiber (8.4%) and ash (4.8%), (Babayan et al., 1978).

In addition, *N. sativa* meal is considered one of these alternatives feeds which rich in secondary compounds e.g. saponins, flavonoids, nigellone, tannins, resin, and volatile oils that could have positive influence on human and animal health. The active compound in *N. sativa* seed has been reported to have therapeutic effect including antioxidant (Kanter et al., 2003) and antimicrobial (Khan et al., 2003). *Nigella sativa* meal contains most of essential amino acid, with high level of minerals (Elmowalid et al., 2013).

The addition of *N. sativa* meal in the diet improved growth performance of broilers (Guler and Ertas, 2006; Abu-Dieyeh and Abu-Darwish, 2008; Islam et al., 2016) and feed conversion and importantly decreasing feed costs and hence improved economic efficiency (Abdel-Magid et al., 2007). A few studies investigated the effect of use of *N. sativa* meal in sheep (Zanouny et al., 2013) and Mahmoud and Bendary (2014) and goat (Habeeb and El Tarabany, 2012) feeding. The level from 1 up 35% of *N. sativa* meal had a positive impact on animal performance (El-Ghammry et al., 2002; Mohamed, 2007; El-Rahman et al., 2011)

In Egypt, ruminant rations still depend mainly on cottonseed cake as a source of protein, however due to inadequate supply with cottonseed cakes in the last few years, it is necessary to search for other oil seed crops that can produce a good quality by product which can substitute the cottonseed cake. The replacement of these meals with alternative meals may be decrease in feed cost. This study aimed to investigate the replacement of traditional source of protein (cotton seed meal) by *N. sativa* meal on growth performance, blood metabolites and rumen fermentation parameters of growing Farafra lambs.

## **MATERIALS AND METHODS**

### ***Animals, diets and management:***

The experimental trail was carried out at the Animal Production Research Farm, Faculty of Agriculture, Assiut University, Egypt. Eighteen of Farafra male lambs (aged 5 to 6 month and had an average of  $27.2 \pm 1.3$ kg body weight divided into three homogenous groups (n=6) for 90 days. The lambs were individually housed in cages (1.2×1.2 m<sup>2</sup>) and assigned randomly to one of three following groups, a control group (CON) fed basal diet of concentrate mixture plus wheat straw and and treated groups (T1 and T2), they were fed basal diet plus 33.3% (T2) and 66.7% (T3) as a substitution of cotton seed meal by *N. sativa* meal in concentrate mixture .

The lambs were fed 65% of their requirements as a concentrate mixture based on NRC guidelines and the rest were covered from wheat straw as roughages *ad libitum*. Lambs were weighed at the beginning of the experiment and over two weeks to adjust the quantity of concentrate mixture. Along the experimental period, the concentrate mixture was offered for each animal individually once daily at 9.00 am. After consumption of the concentrate, wheat straw was added to lambs at 12.00 hr. taking into the consideration the forage to concentrate ratio. Residual feed was weighed in the morning next day at 8.00 h to calculate the individual feed intake. Drinking water was freely available to the lambs in each pen. Feed conversion ratio was calculated and expressed in terms of gram dry matter (DM) per gram body weight gain. The growth experimental periods involved of two periods, 15-days adjustment period followed by 90 days experimental period. The chemical analysis of feeds was carried out using procedures of Association of the Official Analytical Chemists (AOAC, 2012). Ingredients and chemical composition of experimental diets used in this study are shown in Table (1).

### ***Blood sampling***

At the end of the experiment about 10 ml of blood samples were taken from the jugular vein of each lamb in tubes containing potassium ethylene diamine tetra-acetic acid (K-EDTA). The blood samples were directly centrifuged at 3000 rpm for 15 min and the plasma was recovered and stored at -20 °C until analyzed. Plasma metabolites: total protein, albumin (globulin concentration was obtained as the difference between the total protein and albumin concentration) , total cholesterol, triglycerides, alanine aminotransferase (ALT), aspartate aminotransferase (AST) enzymes using assay kits supplied by Diamond, Egypt. Thyroid hormones (T3 and T4) concentrations were determined using enzyme-linked immunosorbent assay kits supplied by Biotecx, USA. The method of (Koracevic et al., 2001) is used to determine the Total antioxidant capacity (TAC).

### ***Digestibility trial:***

The digestibility trials were carried out using twelve local rams (two years old and about  $52 \pm 1.25$  kg body weight). Each digested trial lasted for 21 dayes, the first 15 days were considered as a preliminary period followed by 7 days collection period. Animals were randomly allocated into three experimental groups, each with 4 rams. The animals in group one were considered as a control, fed basal diet of concentrate mixture plus wheat straw and treated groups were substitution 33.3% (T1) or 66.7% (T2) of cotton seed meal in concentrate mixture by *N. sativa* meal.

**Table (1). The ingredients and chemical composition of experimental rations**

Ingredient (% of ration)	Treatments*			Wheat straw	<i>N. sativa</i> meal
	Control	T1	T <sub>2</sub>		
Sorghum	20	20	20	-	
Maize Grain	20	20	20	-	
Wheat Bran	35	35	35	-	
Cotton seed cake meal	21	14	7	-	
<i>Nigella sativa</i> meal	-	7	14	-	
Limestone	2	2	2	-	
Salt	1	1	1	-	
Trace mineral and vitamin premix <sup>‡</sup>	1	1	1	-	
Chemical composition (%)					
DM	90.58	90.54	90.50	95.36	90.35
OM	82.84	83.30	83.76	84.69	81.60
CP	14.02	14.50	14.94	3.26	33.3
EE	2.90	3.42	3.93	1.56	12.75
NDF	29.90	29.22	28.50	72.50	34.96
ADF	12.90	12.23	11.55	43.92	22.00

<sup>‡</sup> The premix consist of (per kg): 20,000,000 IU vitamin A, 10,000 mg vitamin E, vitamin 200,000 IU D3, 10,000 mg Fe, 2500 mg Cu, 100 mg Mo, 20,000 mg Mn, 100 mg Co, 800 mg I, 20,000 mg Zn and 100 mg Se.

\* T1: substitution 33.3% of cotton seed meal by *N. sativa* meal. T2: substitution 66.7% of cotton seed meal by *N. sativa* meal.

#### **Chemical analysis and digestion coefficients measurements:**

During the collection period of feces, the daily feed intake of concentrate mixture and wheat straw was calculated by removal of residual feed from the offered one. Samples of diets were taken, mixed and ground through 1mm screen and stored for chemical analyses. Feces were collected daily and 10% of its weight were taken and dried at 60-70 °C for 24 hrs. The fecal collected samples from each animal were composited and grounded through a 1 mm mill screen for following chemical analysis. The chemical analyses of feed samples, and feces were conducted by using the methods of AOAC (2012). The apparent digestion coefficients of nutrients were defined by expressing the difference between the content of nutrients in both consumed feed and feces as a ratio of its intake.

#### **Rumen fermentation activities**

At the end of digestibility trial, samples of rumen contents were collected once from each ram using a stomach tube. The samples were collected 3-4 hrs after the morning feeding. These rumen liquor samples were divided into two portions, the first portion filtrated through one layer of cheese-cloth, which utilized to measure the protozoal count. The total protozoal count was carried out according to Abou El-Naga (1967). However, the second part filtrated through four layers of cheese-cloth, and then the obtained filtrate was used directly for measurement of pH using a digital pH meter, and ammonia N concentration according to Conway (1962) method. Only Some drops of saturated liquid of mercuric chloride were added to the filtrate to stop the microbial activity before its storage for analysis, and then the samples were kept frozen at -20°C for determination total volatile fatty acids (VFAs). The total VFAs acids were measured using the procedures of Warner (1964).

#### **Statistical Analysis**

The results were statistically analyzed using of SPSS statistical package 22 (SPSS Institute, Chigaco, IL, USA). The data were assessed using the General Linear Models (GLM) procedure for analysis of variance and subjected to one-way ANOVA accompanied by Duncan's multiple range tests to detect the differences among the treatments. The data are presented as means± SE. Probability values less than 0.05 ( $P < 0.05$ ) was considered significant.

The statistical model was as follows:  $Y_{ij} = \mu + T_i + e_{ij}$

Where:  $Y_{ij}$  = the observation  $ij$ ,  $\mu$  = the overall mean,  $T_i$  = the effect due to treatment  $i$ ,  $e_{ij}$  = the experimental error.

## RESULTS AND DISCUSSION

**Lambs growth performance:**

The results in Table (2) indicate that the average body weight gain (BWG) and the average daily gain (ADG) of T2 group were higher ( $P < 0.05$ ) than T1 and CON group. Inclusion of *N. sativa* meal as alternative of cotton seed meal with rate 66.7% to diet of lambs improved average daily gain by about 11.7 and 16.8 % as compared with T1 and control group, respectively. However, there was no difference ( $P > 0.05$ ) between T1 and control group. The positive effect of *N. sativa* meal on growth performance may be due to its content of essential fatty acids like linolenic, oleic acid and linoleic acid that necessary for body growth (Abdel-Azeem et al., 1999; Makkar et al., 2007). Also, Retnani, et al. (2019) found that addition *N. sativa* meal to the diet do it extra palatability for the lambs which caused total dry matter and feed protein intake to be greater than those fed CON diet. According to the NRC (2006), one of the factors that could influence weight gain is the total protein obtained daily from the feed consumed. In addition, this improvement in daily gain in this study may be related to higher digestibility coefficient of almost nutrients especially the increase in CP and total DM intake (Table 4). Similarly, Abd El-Rahman et al. (2011) found that fed 20% *N. sativa* improved ( $P < 0.05$ ) average daily gain of Demeshgi goats compared to the CON diet. Also, Mohamed (2007) found that an improvement of camel performance fed 35% of *N. sativa* substituted control diet protein. Abdel-Magid et al. (2007) stated that substituting 30% or 60% of the crude protein in a feed ration with *N. sativa* meal increased the average daily gain up to 10% compared with control. Also, Cherif et al. (2018) stated that the supplement of *Nigella* seeds improved ( $P < 0.05$ ) the growth performance of the Barbarine lamb.

**Table (2). Effect of substituted cotton seed meal by *N. sativa* meal on growth performance of growing lambs**

Item	Treatment*			P value
	Control	T1	T2	
Initial weight (kg)	27.33 ± 0.33	27.66 ± 0.61	27.33 ± 0.49	0.861
Final weight (kg)	46.33 <sup>b</sup> ± 0.33	47.83 <sup>b</sup> ± 1.60	50.16 <sup>a</sup> ± 0.47	0.001
BW gain (kg)	19.00 <sup>b</sup> ± 0.51	20.16 <sup>b</sup> ± 0.16	22.83 <sup>a</sup> ± 0.45	0.001
Daily gain (g)	211.11 <sup>b</sup> ± 5.73	224.07 <sup>b</sup> ± 1.85	253.70 <sup>a</sup> ± 5.30	0.001
Feed Intake (FI, g/day)				
DMI of concentrate	1358.85 ± 0.00	1297.59 ± 51.83	1297.02 ± 39.90	0.500
DMI of wheat straw	743.99 <sup>b</sup> ± 40.18	713.61 <sup>b</sup> ± 64.38	890.50 <sup>a</sup> ± 10.22	0.029
Total DM intake	2102.84 <sup>ab</sup> ± 40.18	2011.20 <sup>b</sup> ± 55.50	2175.20 <sup>a</sup> ± 36.39	0.043
CP intake	295.03 <sup>b</sup> ± 5.63	291.02 <sup>b</sup> ± 8.03	326.81 <sup>a</sup> ± 5.43	0.003
Feed conversion ratio g/g gain)	9.96	8.98	8.57	

<sup>a, b, c</sup> Means of the same row in each item with different superscripts are significantly different ( $P < 0.05$ ).

\*T1: substitution 33.3% of cotton seed meal by *N. sativa* meal. T2: substitution 66.7% of cotton seed meal by *N. sativa* meal.

The results in Table (2) indicate that the daily DM intake from wheat straw by lambs fed ration T<sub>2</sub> was higher ( $P < 0.05$ ) by about 19.9 and 16.5% than those fed T1 and control groups, respectively. Also, the total DM and CP intake were increased ( $P < 0.05$ ) in T<sub>2</sub> group as compared with control group. The improvement fed intake with increasing the level of substitution cotton seed meal by *N. sativa* meal may be due to *N. sativa* meal have been extensively used as a digestive and appetite stimulant (Cherif et al., 2018). The taste (bitter and pungent) of the *N. sativa* seeds is incurred by their phenolic compounds and essential oils (Aggarwal, 2009). Idris et al. (2014) attributed the improvement fed intake of lambs fed commercial oil of *N. sativa* to their appetizer effect. The similar results were reported by Habeeb and El-Tarabany (2012) they confirmed that the addition of *Nigella* to the diet of Zaraibi kids enhanced ( $P < 0.05$ ) DM intake. Abd El-Ghani (2003) reported that the use of *N. sativa* meal significantly increased the average feed intake and body weight. Similarly, Cherif et al. (2018) noted that the addition of *Nigella* seeds to high concentrate diet enhanced ( $P < 0.05$ ) the total DM, OM, CP and NDF intakes of lambs. However, Retnani, et al. (2019) reported that supplement *N. sativa* meal to the diet of lambs at rate of 10 and 20% did not affected on dry matter (DM) intake.

Regarding feed conversion ratio data presented in Table 2 showed that substitution of cotton seed meal by *N. sativa* meal improved feed conversion ratio by about 10.91 and 16.2% for T<sub>1</sub> and T<sub>2</sub> groups as compared with control, respectively. The positive effects of *N. sativa* meal supplement on average daily gain reflected on feed conversion ratios that are consistent with literature. Cherif et al. (2018) stated that the addition of *Nigella* seeds to high concentrate diet increased (P <0.05) feed conversion ratio than high rough diet with or without *Nigella* seeds supplement.

**Blood metabolites:**

The data in Table (3) revealed that there were no significant differences (P>0.05) in serum total protein, albumin, globulin, A/G ratio, triglycerides, AST and T<sub>3</sub> hormone of lambs due to *N. sativa* meal diet. However, the average value of plasma cholesterol concentration and ALT of lambs fed diets with *N. sativa* meal were lower (P<0.05) than that of lambs fed control diet. Moreover, substituted cotton seed meal by *N. sativa* meal at level 33.4% in T<sub>1</sub> group increase (P<0.05) glucose and T<sub>3</sub> hormone compared with T<sub>2</sub> and control groups. These results were confirmed by Zanouny et al. (2013) they reported that the supplementation of *N. sativa* to lambs diet decreased plasma triglycerides and plasma cholesterol concentration. Retnani, et al. (2019) indicates that the use of *N. sativa* meal in the diet did not alter the blood glucose or blood triglyceride concentrations of the lambs. The lower value of cholesterol may be attributed to the increase of thyroid gland activity (Table 3) as a result of *N. sativa* meal supplementation. Mohammed and Al-Suwaiegh (2016) stated that *N. sativa* can accelerate thyroid gland directly or indirectly via the pituitary gland. Thyroid hormones are very important for the metabolisms which increase the metabolic rate that can lead to enhanced amino acid utilization by fastening their metabolism. Also, Daghash et al. (1999) observed that the addition of *N. sativa* seeds on rabbit diet reduced serum cholesterol, total lipids and triglycerides concentration. The reduction in the triglycerides and cholesterol level might be due to the active ingredients in *N. sativa* such as thymoquinone and compounds like monounsaturated fatty acids that lower the cholesterol production by hepatocytes and reduce the fractional absorption of cholesterol from small intestine (Brunton, 1998). Also, Omar (2003) attributed the reduced cholesterol and triglyceride concentrations in diets containing high levels of *N. sativa* meal to unsaturated fatty acids, which may encourage the cholesterol secretion into the intestine and the oxidation of cholesterol to bile acids.

Transaminases enzymes (AST and ALT) concentrations were not significantly affected by *N. Sativa* (Table 3). The groups of lambs fed rations containing *N. sativa* meal had higher (P<0.05) concentration of plasma antioxidant capacity (TAC) than that of CON group. These antioxidant effects of *N. sativa* seeds might be due to the active constituents like thymoquinone, carvacole, anethole and 4- terepinol (Guler et al., 2007). Badary et al. (2003) concluded that *N. sativa* is an excellent superoxide anion scavenger for free radicals.

**Table (3). Effect of substituted cotton seed meal by *N. sativa* meal in lambs ration on some blood parameters**

Item	Treatment*			P- value
	Control	T1	T2	
Glucose (mg/dl)	46.39 <sup>b</sup> ± 0.69	59.71 <sup>a</sup> ± 1.22	46.91 <sup>b</sup> ± 0.89	0.001
Total protein (g/dl)	5.72 ± 0.13	7.12 ± 0.67	7.04 ± 0.65	0.201
Albumin (A) (g/dl)	2.47 ± 0.08	2.72 ± 0.25	2.73 ± 0.23	0.613
Globulin (G) (g/dl)	3.25 ± 0.10	4.40 ± 0.84	4.31 ± 0.44	0.326
A/G ratio	0.76 ± 0.04	0.69 ± 0.21	0.64 ± 0.04	0.789
Cholesterol(mg/dl)	58.56 <sup>a</sup> ± 2.82	30.67 <sup>b</sup> ± 4.89	29.54 <sup>b</sup> ± 1.49	0.001
Triglycerides (mg/dL)	177.36 ± 4.96	160.18 ± 4.87	159.64 ± 6.41	0.086
ALT(U/l)	15.12 <sup>a</sup> ± 1.65	10.75 <sup>b</sup> ± 0.24	7.63 <sup>b</sup> ± 0.61	0.015
AST(U/l)	36.78 ± 0.76	39.29 ± 7.76	36.88 ± 6.93	0.946
T3 (ng/ml)	2.85 <sup>b</sup> ± 0.08	3.58 <sup>a</sup> ± 0.01	3.27 <sup>ab</sup> ± 0.03	0.071
T4 (µg/dl)	15.33 ± 1.02	17.03 ± 0.09	15.73 ± 0.92	0.350
TAC (mmol/L)	0.75 <sup>b</sup> ± 0.01	0.88 <sup>a</sup> ± 0.01	0.93 <sup>a</sup> ± 0.02	0.001

<sup>a, b, c</sup> Means of the same row in each item with different superscripts are significantly different (P<0.05).

\*T1: substitution 33.3% of cotton seed meal by *N. sativa* meal. T2: substitution 66.7% of cotton seed meal by *N. sativa* meal.

**Nutrient digestibility coefficients**

Inclusion of *N. sativa* meal as partially alternative of cotton seed meal to diet of lambs improved ( $P < 0.05$ ) nutrients digestibility as compared with control group (Table 4). Also, it could be noticed that the nutrient digestibility was increased ( $P < 0.05$ ) with increasing substitution rate from 33.3 to 66.7% for cotton seed meal by *N. sativa* meal. *Nigella sativa* has antibacterial characteristics (Bita et al., 2012). Therefore, *Nigella sativa* may modulate microorganisms of rumen and their capabilities of rumen digestion. Lewis (1980) reported that the addition of *N. sativa* in feed heightened bile flow rate results in enhanced emulsification that activates the pancreatic lipases, which then aid in fat digestion and absorption of fat soluble vitamins. Jamroz and Kamel (2002) noticed a stimulating effect of black seeds on digestive system, resulting in superior absorption and performance. The results of our study agreement with finding by Abdel-Magid et al. (2007) found that substitution 30% or 60% of soybean meal by *N. sativa* meal in growing calves rations showed specific improvement in nutrients digestibility. Aslo, Retnani, et al. (2019) found that addition *N. sativa* meal to the diet of lambs at rate of 10 and 20% improved the digestibility of crude protein. Similarly, Khattab et al. (2011) informed that calves fed ration added with black seed oil had better nutrient digestibility compared to calves fed free black seed oil diet. However, Cherif et al. (2018) found that the enrichment diets of lambs with *Nigella* seeds did not affect ( $P < 0.05$ ) the apparent digestibility of DM, OM, CP and NDF of these diets. Also, Awadalla (1997) found that replacing sunflower meal-protein at 0, 50 and 100% by black cumin seed-meal in the concentrate mixture of growing sheep not significantly effected on nutrient digestibility.

**Table (4). Effect of substituted cotton seed meal by *N. sativa* meal in rams ration on nutrient digestibility, %**

Item	Treatment			P- value
	Control	T1	T2	
Dry matter, DM	64.35 <sup>c</sup> ± 1.81	70.23 <sup>b</sup> ± 1.05	76.51 <sup>a</sup> ± 0.87	0.005
Organic matter, OM	65.11 <sup>c</sup> ± 1.05	68.80 <sup>b</sup> ± 0.88	76.46 <sup>a</sup> ± 0.75	0.003
Crude protein, CP	70.23 <sup>b</sup> ± 1.04	73.75 <sup>ab</sup> ± 0.99	77.00 <sup>a</sup> ± 1.08	0.010
Ether Extract, EE	66.95 <sup>c</sup> ± 0.18	70.75 <sup>b</sup> ± 1.08	75.72 <sup>a</sup> ± 1.40	0.002
Crude fiber, CF	54.28 <sup>b</sup> ± 0.96	56.69 <sup>ab</sup> ± 0.44	58.46 <sup>a</sup> ± 0.74	0.020
Nitrogen free extract, NFE	79.90 ± 0.92	80.14 ± 1.92	79.33 ± 1.44	0.924

<sup>a, b, c</sup> Means of the same row in each item with different superscripts are significantly different ( $P < 0.05$ ).

\*T1: substitution 33.3% of cotton seed meal by *N. sativa* meal. T2: substitution 66.7% of cotton seed meal by *N. sativa* meal.

**Rumen fermentation activities**

As represented in Table 3, partially substitution of cotton seed meal by *N. sativa* meal not affected on rumen pH and was maintained at 6.2 to 6.4 in all dietary treatment groups. However, ruminal concentrations of NH<sub>3</sub>-N and total VFAs increased ( $P < 0.05$ ) with increasing substitution level from 33.3 to 66.7% for cotton seed meal by *N. sativa* meal as compared with control group. The higher concentration of NH<sub>3</sub>-N in rumen of lambs receiving *N. sativa* meal may be associated with increasing CP intake (Table 2) or CP digestibility (Table 4). While, the difference of the concentration of NH<sub>3</sub>-N in the rumen of sheep between groups taking *N. sativa* meal could be the result of the effect of *p-cymene* which is the major element of oil in *Nigella* seeds (Chaves et al., 2008). Busquet et al. (2006) stated that *p-cymene* stimulates the microbial deamination activity. In addition, the *p-cymene* in *Nigella* oil has an inhibitory effect on bacterial peptidolysis and deamination processes. Ali (2003) found that the concentration of rumen ammonia nitrogen increase with increasing amounts of *N. sativa* meal in the diets of lambs. In the same context, Cherif et al. (2018) reported that the administration of *Nigella* seeds reduced ( $P < 0.01$ ) the total number of protozoa in the rumen, but it increased ( $P < 0.01$ ) the concentration of NH<sub>3</sub>-N.

The higher value of total VFAs for animals fed *N. sativa* meal in T1 and T2 are in agreement with El-Naggar et al. (2018) and Klevenhusen et al. (2015) they found that, the rumen volatile fatty acids increased ( $P < 0.05$ ) with increasing the level of *N. sativa* oil of lambs ration from 0.1 and 0.2% as compared with control group.

The total number of protozoa was decreased ( $P < 0.05$ ) gradually with increasing the level of *N. sativa* meal in the diets of lambs (4.70 and 2.24 vs.  $1.20 \times 10^6/\text{ml}$ ) for T<sub>1</sub>, T<sub>2</sub> and CON group, respectively. The smaller number of protozoa in lambs receiving *N. sativa* meal may be due to some ingredients in *Nigella* such as thymoquinone and thymohydroquinone and these ingredients possess antimicrobial properties (Mohammed and Al-Suwaiegh, 2016). This results agreement with Cherif et al. (2018) they reported that the protozoa population size decreased with addition of *N. sativa* meal to the diets of lambs.

**Table (5). Effect of substituted cotton seed meal by *N. sativa* meal in lambs ration on rumen parameters**

Item	Treatments*			P-value
	Control	T1	T2	
pH	6.28 ± 0.03	6.32 ± 0.11	6.44 ± 0.04	0.318
Ammonia, mg/100 ml	14.50 <sup>c</sup> ± 0.44	17.77 <sup>b</sup> ± 0.64	23.69 <sup>a</sup> ± 1.12	0.005
TVFA, mmol/100 ml	7.30 <sup>c</sup> ± 0.50	8.93 <sup>b</sup> ± 0.39	11.47 <sup>a</sup> ± 0.51	0.002
Total protozoa count, ×10 <sup>6</sup> /ml	4.70 <sup>a</sup> ±0.177	2.24 <sup>b</sup> ±0.034	1.20 <sup>b</sup> ±0.023	0.002

<sup>a, b, c</sup> Means of the same row in each item with different superscripts are significantly different ( $P < 0.05$ ).

\*T1: substitution 33.3% of cotton seed meal by *N. sativa* meal. T2: substitution 66.7% of cotton seed meal by *N. sativa* meal.

## CONCLUSION

Inclusion of *N. sativa* meal as partially substitution of cotton seed meal to diet of lambs improved the body weight, the average daily gain, nutrients digestibility and rumen fermentation activity of growing Farafra lambs. From the previous results it could be concluded that addition of *N. sativa* meal until rate 66.7% as alternative source for cotton seed meal in diets of sheep may recommended to improve feed utilization and growth performance of growing Farafra sheep.

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## تأثير الاستبدال الجزئي لكسب بذرة القطن بكسب حبة البركة على هضم العناصر الغذائية، تخمرات الكرش، بعض خصائص الدم و أداء الحملان النامية

محمود عبد اللطيف محمد عبد الله و محسن محمد فرغلى

قسم الانتاج الحيوانى- كلية الزراعة – جامعة أسيوط - مصر

أجريت هذه الدراسة لتقييم تأثير الإستبدال الجزئى لكسب بذرة القطن فى المخلوط المركز بكسب حبة البركة على هضم العناصر الغذائية، تخمرات الكرش وخصائص الدم وأداء الحملان النامية. وتم استخدام 18 حمل من اغنام الفرازة عمر 5-6 شهور و بمتوسط وزن  $27.2 \pm 1.3$  كجم قسمت بشكل عشوائى الى ثلاثة مجاميع متجانسة ( 6 حملان فى المجموعة) لمدة 90 يوم. وكانت المجاميع عبارة عن مجموعة الكنترول وتم تغذيتها على المخلوط المركز مع تين القمح لحد الشبع، مجاميع المعاملة وفيها تم استبدال نسبة (33.3%) من كسب القطن فى المخلوط المركز بكسب حبة البركة مع تعذيه تين القمح حتى الشبع فى حين المعاملة الثانية تم استبدال نسبة (66.7%) من كسب القطن فى المخلوط المركز بكسب حبة البركة مع تعذيه تين القمح لحد الشبع. تم تغطيه 65 % من احتياجات الحملان من العليقة المركزة طبقا للـ NRC فى حين بقيه الاحتياجات تم تغطيتها من تين القمح. وتم اجراء ثلاث تجارب هضمهم باستخدام 12 كبش تام النمو وزعت بشكل عشوائى فى صناديق الهضم وتم تغذيتها على نفس العلائق السابقة. فى تجربة النمو تم تسجيل ومتابعة أوزان الحيوانات وحساب معدل نموها وتم أخذ عينات دم لتقدير انزيمات الكبد والغدة الدرقية وبعض مكونات الدم ومضادات الاكسدة الكلية. وفى تجارب الهضم تم تجميع عينات كرش لتقدير درجة الحموضة وتركيز الأمونيا والاحماض الدهنية الطيارة الكلية وأعداد البروتوزوا وتم أيضاً تحليل عينات العلف الروث. وتشير النتائج الى هناك زيادة معنوية فى الوزن النهائى والزيادة اليومية فى مجموعة المعاملة الثانية مقارنة بمجموعة المعاملة الاولى والكنترول. زاد معدل الغذاء اليومي الماكول فى صورة مادة جافه و بروتين خام بشكل معنوي فى المعامله الثانيه مقارنة بمجموعه المقارنه. الاستبدال الجزئى لكسب القطن بكسب حبه البركه ادى الى زيادة معنويه فى تركيز الامونيا والاحماض الدهنيه الطيارة فى الكرش مع نقص فى اعداد البروتوزوا فى مجموعات المعاملات بحبه البركة بالمقارنة بالمجموعة الكنترول. ايضا كان هناك انخفاض فى تركيز معنوى الكوليستيرول وانزيم الألتين أمينوترانسفيراز ALT فى مجموعتى كسب حبة البركة مقارنة بالمجموعة الكنترول. بينما ارتفع مستوى هرمون الغدة الدقية T3 و مستوى مضادات الاكسدة فى مجموعتى حبة البركة مقارنة بالمجموعة الكنترول. ومن تجربة الهضم اتضح ان هناك زيادة معنوية فى هضم مع زيادة العناصر الغذائية المختلفة نسبة أستبدال كسب القطن بكسب حبة البركة ومن تجربة الهضم اتضح ان هناك زيادة معنوية فى هضم العناصر الغذائية المختلفة مع زيادة نسبة أستبدال كسب القطن بكسب حبة البركة. لذلك فان هذه الدراسه توصى بإمكانيه استبدال جزء من كسب القطن فى علائق الحملان الناميه بكسب حبة البركة كمصدر للبروتين غير تقليدى دون حدوث أى تأثيرات سلبية على أداء الحملان الناميه.

**الكلمات المفتاحية:** كسب حبة البركة ، هضم العناصر الغذائية، تخمرات الكرش، خصائص الدم ، أداء الحملان.