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Effect of Partial Substitution of Dietary Protein by *Nigella sativa* Meal and Sesame Seed Meal on Performance of Egyptian Lactating Buffaloes

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ABSTRACT

This study aimed to find out the impact of partial substitution of dietary protein by *Nigella sativa* meal (NSM) and Sesame Seed Meal (SSM) on milk yield and composition, digestibility, some of blood parameters as well as economic efficiency. Sixteen lactating multiparous Egyptian buffaloes were used 8 weeks after calving. They were assigned into four groups (4 buffaloes each) to be fed on one of the four experimental rations, each ration consisted of 50% roughages plus 50% of the different concentrate feed mixtures differ in protein source, in 4×4 Latin square design. The experimental rations were as follow, R1: Traditional concentrate feed mixture plus Clover (C) plus Wheat Straw (WS), R2: Concentrate feed mixtures contained 50% NSM as protein source plus C plus WS, R3: Concentrate feed mixtures contained 50% SSM as protein source plus C plus WS and R4: Concentrate feed mixtures contained 25% NSM and 25% SSM as protein source plus C plus WS. No significant ($p < 0.05$) differences among rations in the digestibility of DM, CF, NFE, NDF, cellulose and hemi-cellulose were observed, while, OM, CP, EE and ADF showed significant differences among rations. The 3rd ration recorded the highest TDN (61.79%) followed by control ration (56.61%) however R2 and R3 showed intermediate values. The highest 7% FCM was recorded with buffaloes fed R3 (11.01 kg h⁻¹ day⁻¹) followed by feeding R4, R1 and R2 being 10.56, 10.52 and 10.08 kg h⁻¹ day⁻¹, respectively. In general, the present results indicate that *Nigella sativa* meal and sesame meal were suitable for feeding lactating buffaloes and formulated balanced rations with adequate protein and energy which will be reflected on good health condition and normal performance of all lactating buffaloes.

Key words: Buffaloes, milk, *Nigella sativa* meal, sesame seed meal, nutrients digestibility

INTRODUCTION

The sustainability of large animal projects development is absolutely dependent on the availability of quality and cheap feedstuffs. Plant meals represent the main source of protein in ruminant rations. The protein level in Egyptian animal rations is usually accomplished by feeding cotton seed meal and soybean meal. Cottonseed meal (CSM) is a source of rumen by-pass protein supplement in dairy cattle feeding (Wanapat *et al.*, 1996). Several studies have been devoted to study the potential value of CSM in ruminants (Erdman *et al.*, 1987; Da Cunha *et al.*, 1998; Sampaio *et al.*, 2000). On the other hand, use of un-decorticated cottonseed meal as an animal feed is limited by the presence of gossypol and decreasing cultivated area of cotton. Soybean meal (SBM) is an important ingredient in many countries which has a high crude protein (CP) content (43.7-48% DM) (Castillo *et al.*, 2001). It provides a combination of amino acids that can

support high milk yields (McDonald *et al.*, 2002). However, local conditions for soybean cultivation are not always favorable and it is expensive to import SBM. *Nigella sativa* are cultivated in the Mediterranean region and Asia (Hutchinson, 1959). These seeds are rich in oil (Abdel-Aal and Attia, 1993a) and the defatted black seeds (black seed cake) has a high level of true protein (Abdel-Aal and Attia, 1993b). The meal contains most of the essential amino acids, with crude protein about 33% (El-Nattat and El-Kady, 2007). Sesame (*Sesamum indicum* L.) seed is a drought-tolerant crop adapted to many soil types (Ram *et al.*, 1990). Sesame seed meal after oil extraction is excellent source of edible nutrients (45-50% lipid, 15-20% protein and 10-15% carbohydrate (Lee *et al.*, 2005) and 47.1-52.9% CP (Mamputu and Buhr, 1995; Kaneko *et al.*, 2002). Previous data indicated that providing different sources of protein like *Nigella sativa* meal and sesame seed meal in ruminant rations are important matter. Therefore, this study aimed to find out the impact of partial substitution of dietary protein by *Nigella sativa* meal and sesame seed meal on milk yield and composition, digestibility, some of blood parameters as well as economic efficiency.

MATERIALS AND METHODS

Experimental animals and rations: Sixteen lactating multiparous Egyptian buffaloes (in 2nd to 5th lactation season) weighed 590 kg on average were used after 8 weeks of calving, they were assigned into four groups of 4 buffaloes. Each group of buffaloes were fed one of the four experimental rations, each ration consisted of 50% roughages plus 50% one of different concentrate feed mixture (CFM) in 4×4 Latin square design. Each period was divided into 21 days for adaptation and 7 days for samples collection. The first CFM contained soybean meal (SBM) and cotton seed meal (CSM) as the common source of protein in Egypt, while 50% of SBM and CSM were replaced by *Nigella sativa* meal (NSM) or sesame seed meal (SSM) or by both tested meals in 2nd, 3rd and 4th CFM's as shown in Table 1.

Feeding procedures: Animals were individually fed on the experimental rations to cover total requirements from energy and protein according to Ghoneim (1964). The CFM and clover (*Trifolium alexandrinum*) and wheat straw were offered to all animals after milking time (7.00 am and 7.00 pm). Water was available at all times.

Table 1: Formulation of the experimental concentrate feed mixtures (CFM)

Item	Concentrate Feed Mixtures (CFM)			
	CFM1	CFM2	CFM3	CFM4
Yellow corn	50.0	50.0	50.0	50.00
Wheat bran	12.0	12.0	12.0	12.00
Cotton seed meal	23.0	11.5	11.5	11.50
Soybean meal	12.0	6.0	6.0	6.00
<i>Nigella sativa</i> meal	-	17.5	-	8.75
Sesame meal	-	-	17.5	8.75
Mineral mix*	1.0	1.0	1.0	1.00
Salt	0.5	0.5	0.5	0.50
Limestone	1.5	1.5	1.5	1.50

*Mineral mix: 50 g Zn, 50 g Mn, 50 g Fe, 10 g Cu, 0.50 g I, 0.10 g Co and 0.20 g Se plus CaCO₃ up to 2 kg

Milk recording: Buffaloes milk was obtained by machinery milking two times daily (7.00 am and 7.00 pm) and the daily milk yield was individually recorded and was corrected to 7% FCM by the following equation according to Raafat and Saleh (1962):

$$7\% \text{ FCM} = (0.265 \times \text{milk yield}) + (10.5 \times \text{fat yield})$$

Milk sampling and analysis: During the 7 days collection period, two milk samples were individually collected from each buffaloes at 7.00 am and 7.00 pm and were composite. Chemical analysis of milk (fat, protein, solids not fat, total solids and ash) was determined according to Ling (1963). Lactose was calculated by difference.

Digestibility trials: At the end of each collection period, nutrients digestibility were determined by the Acid Insoluble Ash (AIA) technique as described by Van Keulen and Young (1977) to determine the nutrients digestibility and the nutritive values of the experimental rations. Feces samples were collected for six successive days from each animal. Total digestible nutrients (TDN) and digestible crude protein (DCP) were calculated according to McDonald *et al.* (1995).

Chemical analysis: Feed ingredients, CFM, roughage and feces were analyzed for proximate analyses according to AOAC (2000). Nitrogen free extract was calculated by difference. Fiber fractions were analyzed according to Van Soest *et al.* (1991). Hemi-cellulose and cellulose were calculated as follows:

$$\text{Hemi-cellulose} = \text{NDF} - \text{ADF}$$

$$\text{Cellulose} = \text{ADF} - \text{ADL}$$

Blood parameters: At the end of tested period of each ration after four hours morning feeding blood samples were collected from all the experimental buffaloes. The blood samples were taken from the jugular vein in dry clean glasses tubes using heparin as anticoagulant and then centrifuged for 15 min at 4000 rpm to obtain plasma. Biochemical of blood plasma constituents was determined by using commercial kits, total protein and creatinine as described by Tietz (1986) and Tietz *et al.* (1990), albumin was determined according to Doumas *et al.* (1971), blood plasma urea was determined according to Patton and Grouch (1977). Alanin amino transferase (ALT) and activity of aspartate transfearse (AST) were determined by the methods of Young (1997). Triglyceride was determined according to Fassati and Prencipe (1982) and cholesterol according to Richmond (1973).

Statistical analysis: Data was analyzed using the general linear model procedure of SAS (1999). One way ANOVA procedure used to analyze the intake, digestibility, feed intake, milk production and blood parameter data following the next model:

$$y_i = \mu + T_i + E_i$$

where, μ is the overall mean of y_i , T_i is the ration effect, E_i is the experimental error. The differences among means were separated according to Duncan's New Multiple Range Test (Duncan, 1955).

RESULTS AND DISCUSSION

Chemical composition: Results of chemical composition and fiber fractions of feed ingredients, experimental concentrate feed mixtures and the experimental rations are shown in Table 2, indicated that the chemical composition of *Nigella sativa* meal (NSM) and sesame seed meal (SSM) contained the same proportions of nutrients almost with slight increase of EE in SSM. However, chemical analysis of different concentrate feed mixtures were much closed. Also, fiber fractions constituents were approximately the same with 3% increase in hemi-cellulose content in R4 compared with R1. Generally, *Nigella sativa* meal and sesame meal did not extremely affect on the chemical composition of the experimental rations. Similar results indicating that black cumin seed meal can be a good source of crude protein have been investigated by Awadalla (1997), El-Kady *et al.* (2001) and Abdel-Magid *et al.* (2007). This emphasizes the role that black cumin seed meal can be a rich supplement in animal feeding.

Nutrients digestibility and nutritive values: Data in Table 3, showed that there was no significant ($p < 0.05$) difference among rations in the digestibility of DM, CF, NFE, NDF, cellulose and hemi-cellulose. These results might be due to similar contents of the experimental rations from DM, CF, NFE, NDF, cellulose and hemi-cellulose. While, there were insignificant ($p < 0.05$) decreases in the digestibility of OM, CP and ADF for R2 and R3 but the same nutrients digestibility were significantly ($p < 0.05$) decreased for R4 compared with R1. On the other hand, the digestibility of EE was significantly ($p < 0.05$) higher for R3 than R1 being, 83.78 and 74.77%, respectively. This result might be due to R3 and R4 contained sesame meal which has higher EE content (15.45). There was insignificant ($p < 0.05$) increase in the digestibility of EE for R2 and R4 compared with R1. There was insignificant ($p < 0.05$) difference in TDN among R2, R3 and R4 compared with R1 and the highest value (61.79%) was recorded for R3. These results might be attributed to the highest EE content and its digestibility of R3 (5.51 and 83.78%) compared to other tested ration.

Table 2: Chemical composition of feed ingredients, experimental concentrate feed mixtures and experimental rations (DM basis)

Chemical composition (%)												
Item	DM	OM	Ash	CP	EE	CF	NFE	NDF	ADF	ADL	Cellulose	Hemi-cellulose
Feed ingredients												
C	92.08	88.97	11.03	14.16	1.68	26.60	46.53	46.88	38.18	9.83	28.35	8.70
WS	94.56	83.52	16.48	3.42	1.71	38.59	39.80	77.96	59.49	10.00	49.49	18.47
NSM	90.35	91.55	8.45	33.13	12.72	10.96	34.74	35.30	22.32	4.20	18.12	12.98
SSM	93.85	89.80	10.20	32.35	15.45	9.00	33.00	37.50	24.25	3.50	20.75	13.25
Experimental concentrate feed mixtures												
CFM1	93.59	88.97	11.03	16.73	3.95	6.15	62.14	29.26	14.19	5.68	8.51	15.07
CFM2	92.05	90.01	9.99	16.74	5.21	5.01	63.05	23.95	6.42	4.59	1.83	17.53
CFM3	93.15	92.88	7.12	16.09	10.23	7.04	59.52	28.73	9.87	4.87	5.00	18.86
CFM4	92.41	92.98	7.02	16.15	7.10	7.53	62.20	32.18	9.00	7.24	1.76	23.18
Experimental rations												
R1	93.21	89.23	10.77	13.20	3.11	18.36	54.56	45.26	31.93	8.07	23.86	13.33
R2	92.59	88.27	11.73	13.02	3.24	19.66	52.35	43.39	28.74	7.56	21.18	14.65
R3	93.06	89.74	10.26	13.13	5.51	20.00	51.10	44.31	29.33	7.65	21.68	14.98
R4	92.69	89.53	10.47	13.89	3.86	20.27	51.51	46.96	30.59	8.83	42.76	16.37

C: Clover, WS: Wheat straw, NSM: *Nigella sativa* meal, SSM: Sesame seed meal, CFM1: Traditional concentrate feed mixture, CFM2: NSM concentrate fed mixture, CFM3: SSM concentrate fed mixture CFM4: (NSM + SSM) concentrate fed mixture, R1: CFM1 + C + WS, R2: CFM2 + C + WS, R3: CFM3 + C + WS and R4: CFM4 + C + WS

Table 3: Effect of experimental rations on nutrients digestibility and nutritive values (%)

Item	Experimental rations				±SEM
	R1	R2	R3	R4	
Digestibility (%)					
DM	66.08	64.67	64.00	60.93	2.58
OM	70.17 ^a	68.32 ^{ab}	68.24 ^{ab}	64.58 ^b	2.57
CP	72.28 ^a	66.15 ^{ab}	68.53 ^{ab}	65.42 ^b	2.84
EE	74.77 ^b	79.40 ^{ab}	83.78 ^a	77.11 ^{ab}	3.68
CF	57.09	61.59	60.62	57.37	2.88
NFE	60.34	57.12	59.17	53.32	3.55
NDF	64.69	61.87	62.51	61.55	2.46
ADF	60.39 ^a	55.05 ^{ab}	53.91 ^{ab}	53.23 ^b	3.35
Cellulose	76.89	78.01	79.16	76.72	1.81
Hemi-cellulose	58.44	63.24	64.51	63.32	5.65
Nutritive values (%)					
TDN	56.61 ^{ab}	55.42 ^b	61.79 ^a	54.88 ^b	3.05
DCP	9.54 ^a	8.61 ^b	9.00 ^{ab}	9.09 ^{ab}	0.37

Means in the same row with different superscript are significantly different (p<0.05)

Table 4: Effect of the experimental rations on milk yield and milk composition

Item	Experimental rations				±SEM
	R1	R2	R3	R4	
Milk yield (kg h⁻¹ day⁻¹)					
Actual milk yield	10.40	9.87	10.21	10.70	1.27
7% FCM	10.52	10.08	11.01	10.59	1.32
Milk composition (%)					
Fat	7.11	7.20	7.74	6.90	0.75
Protein	3.25	3.24	3.39	3.29	0.15
Lactose	5.26	5.34	4.98	5.22	0.22
TS	15.62	15.78	16.11	15.41	0.72
SNF	8.51	8.58	8.37	8.51	0.84
Ash	1.03	1.08	1.27	1.13	0.10

Means in the same row with different superscript are significantly different (p<0.05)

Feeding buffaloes R3 or R4 insignificantly (p<0.05) decreased DCP compared with those fed R1 being, 9.00, 9.09 and 9.54%, respectively. While, feeding buffaloes R2 significantly (p<0.05) decreased DCP compared with R1. These results might be due to protein content of R2 was the lowest one among experimental rations. Abdel-Magid *et al.* (2007) and Abd El-Rahman *et al.* (2011) found that substitution 30 or 60% from CFM (5.3 and 10.6% from ration) of soybean meal protein, by black seed did not effect on DM, OM, CP and CF digestibility of rations and in their TDN when evaluated at the end of the experiment. Also, Fitwi and Tadesse (2013) noticed the same results when growing sheep fed on sesame cake from 0, 150, 200, 250 and 300 g day⁻¹.

Milk yield and milk composition: Data in Table 4, indicated that there were insignificant (p<0.05) differences among rations either in milk yield or milk composition. The highest 7% FCM (10.52 kg h⁻¹ day⁻¹) was recorded with buffaloes fed R1. While, feeding buffaloes on R3 recorded the highest fat percentage (7.74%) followed by those fed R2 then R1 and R4 being, 7.20, 7.11 and 6.90%, respectively.

Table 5: Effect of the experimental rations on blood parameters

Item	Experimental rations				±SEM
	R1	R2	R3	R4	
Total proteins (g dL ⁻¹)	7.42	7.80	7.59	7.85	0.31
Albumin (g dL ⁻¹)	5.39	5.56	5.40	5.80	0.27
Globulin (g dL ⁻¹)	2.03	2.24	2.19	2.05	0.14
Urea (mg dL ⁻¹)	23.50	22.50	22.75	21.25	1.19
Creatinine (mg dL ⁻¹)	1.12	1.10	1.07	1.07	0.06
Cholesterol (mg dL ⁻¹)	99.75	113.25	109.50	110.25	8.68
Triglycerides (mg dL ⁻¹)	81.25	79.25	71.25	76.50	7.35
ALT (IU L ⁻¹)	84.50	77.00	101.25	66.00	16.74
AST (IU L ⁻¹)	62.00	66.25	78.50	56.25	11.39

Means in the same row with different superscript are significantly different (p<0.05)

These was expected since the buffaloes in all groups received their recommended nutrients allowances (Ghoneim, 1964). Meantime, all tested rations were contained the same roughage, with the same roughage concentrate ratio. However, the chemical composition of the experimental rations were nearly similar as shown in Table 2. Jabbar *et al.* (2008) stated that partial or complete replacement of cotton seed cake with sunflower meal did not affect daily milk production in the crossbred cattle. There were insignificant (p<0.05) differences among animals fed all tested rations in milk composition. Only R3 which animal fed ration contained sesame meal showed slight insignificant increase with fat, protein, ash and total solid of milk. These results agreement with Kitessa *et al.* (2003), Mustafa *et al.* (2003) and Sarrazin *et al.* (2004) where oil seeds had no or negative effects on ewes milk fat and cows milk fat. On contrast, Zhang *et al.* (2006) and Hejazy (2008) noticed that sesame oil cake increased milk fat percentage. Generally, average of fat, protein, TS, SNF, lactose and ash produced in the present study are within the normal range of milk for buffaloes fed traditional Egyptian ration (Mahmoud and Ebeid, 2014).

Blood parameters: Data related to blood parameters presented in Table 5, did not show any significant (p<0.05) variations among buffaloes fed R2, R3 or R4 compared with those fed R1. Also, results indicated that all parameters were within the normal range according to (Merck, 2014). Total protein, albumin, globulin and urea cleared that no adverse effect of *Nigella sativa* meal and sesame meal on protein metabolism. Plasma proteins concentration can be used as indicator to evaluate the ruminant nutrition during feeding either adequate or low protein levels (Kumar *et al.*, 1980). Kancko (1989) reported that normal range of creatinine in the blood plasma being 1-2 mg dL⁻¹ and protein being 6-8 g dL⁻¹. On the other side, cholesterol and triglycerides observed no bad effect of oil in tested meals on lipid metabolism especially with sesame meal ration.

Feed conversion efficiency and economical evaluation: Data in Table 6, indicated that the intake of dry matter (DM), total digestible nutrients (TDN) and digestible crude protein (DCP) were not influenced by the partial replacement of soybean meal and cotton seed meal by *Nigella sativa* meal, sesame meal and mixtures of tested meals in the ration (p>0.05). Among the factors that may affect DM intake, those that could be influenced by the replacement would be palatability and composition.

Data of economic evaluation presented in Table 6 indicated that there was a decrease in the feed cost per 1 kg 7% FCM with feeding R3 or R4 compared with feeding R1 and R2 being 2.23,

Table 6: Feed intake, feed conversion and economic efficiency of the experimental rations

Item	Experimental rations				±SEM
	R1	R2	R3	R4	
7% FCM	10.52	10.08	11.01	10.59	1.32
Feed intake (kg h⁻¹ day⁻¹)					
CFM	6.00	6.00	6.25	5.50	0.67
Clover	27.50	25.00	27.50	27.50	1.02
Wheat straw	3.12	3.37	2.87	2.87	0.66
Feed intake (h day⁻¹ DM basis)					
DMI (kg)	13.71	13.43	13.71	12.54	1.01
TDN (kg)	7.76	7.45	8.47	6.88	0.58
DCP (kg)	1.31	1.26	1.30	1.14	0.09
Feed conversion ratio					
DM (kg 1 kg 7% FCM)	1.20	1.24	1.41	1.43	0.16
TDN (kg)	0.68	0.69	0.79	0.87	0.09
DCP (kg)	0.11	0.11	0.13	0.13	0.01
Economic study					
Milk price (LE h ⁻¹ day ⁻¹)	63.12	60.48	66.06	63.54	-
Feed cost (LE h ⁻¹ day ⁻¹)	24.93	23.81	24.52	21.73	-
Feed cost /kg 7% FCM, LE	2.37	2.36	2.23	2.05	-
Economic efficiency	2.53	2.54	2.69	2.92	-

Prices (LE: Egyptian pound): CFM 1 (2.34 LE kg⁻¹) CFM 2, 3 and 4 (2.23 LE kg⁻¹), wheat straw (0.8 LE kg⁻¹), Clover (0.22 LE kg⁻¹), milk (6 LE kg⁻¹). Means in the same row with different superscript are significantly different (p<0.05)

2.05 vs. 2.36 and 2.37, respectively. These mainly due to the highest 7% FCM produced by buffaloes fed R3 and R4 compared with those fed other tested rations. Furthermore, economical efficiency as the ratio between price of 7% FCM produced/costs of feed intake showed increased by 6.30 and 10.40% with substituting 50% of SBM and CSM in control ration by SSM or NSM and SSM in 3rd and 4th rations, respectively. Economic efficiency of lactating buffaloes fed R1, R2, R3 and R4 were 2.53, 2.54, 2.69 and 2.92, respectively. In the same context, El-Ayek *et al.* (1999) found that *N. sativa* meal could participate successfully and economically by about 50% of protein in formulating concentrate feed mixture in diets of growing lambs without any adverse effects on animal performance. Fitwi and Tadesse (2013) reported that, using 300 g DM of sesame seed cake was potentially more feasible and economically beneficial for growing sheep.

CONCLUSION

In general, the present results indicate that *Nigella sativa* meal and sesame seed meal are suitable for feeding lactating buffaloes and formulated balanced rations with adequate protein and energy which was reflected on good health condition and normal performance of lactating buffaloes.

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