Effects of Crushed Linseed or Linseed Oil Supplementation on Performance of Dairy Goats and Fatty Acid Profile in Milk

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Abstract: In a complete random block design, fifteen lactating Damascus goats (43.5±2 kg body weight) after the first week of lactation, were divided into 3 groups (five goats each) to evaluate effect of linseed or linseed oil as diet supplement on rumen parameters, milk production, milk composition and milk fatty acid profile in lactating goats. Animals were fed on a total mixed ration of 50% concentrates and 50% berseem clover (control), control ration+50g/head/day crushed linseed (LS) and control ration+20 ml/head/day linseed oil (LO) for 90 days. Dry matter intake was not affected by LO or LS. Ruminal volatile fatty acids (VFA) and butyrate proportions were increased (P<0.05); however, ammonia-N concentration were decreased (P>0.05) with experimental additives. Milk yield, milk protein and milk fat percent were higher (P<0.05) for animals fed LO followed by LS and then control. While, milk urea nitrogen was decreased (P<0.05) with additives. Both od LS and LO decreased (P<0.05) the total saturated fatty acids and increased (P>0.05) the proportion of C18:3N3 in milk fat. Total unsaturated fatty acids were increased (P<0.05) with LO; however, insignificantly increased with LS *versus* control. The proportions of conjugated linoleic acid increased (P<0.05) with addition of LO or LS in the diet. It may be concluded that adding the linseed or linseed oil to lactating goat rations improved the productivity of lactating goats and enhance milk components with no deleterious effects on general health.
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1. Introduction

Feed additives still a new strategy to improve ruminal fermentation (Elghandour *et al*., 2014; Salem *et al*., 2014a, b) and animal performance (Cedillo *et al*., 2014; Valdes *et al*., 2015). The inclusion of saturated fats of animal origin in human diets may increase the risk for cardiovascular diseases (Joyce *et al*., 2009). It has been estimated that dairy products contribute up to 60% of saturated fatty acids (SFA) to human diets (Chilliard *et al*., 2007). Oilseeds are rich in polyunsaturated fatty acids (PUFA), which can be fed to dairy animals to modify the milk fatty acid (FA) profile and produce a more nutritionally beneficial milk for human consumption (Kennelly, 1996). Supplementing ruminant diets with linseeds results in enhanced FA profiles in meat and milk products, that will benefit human health. Moreover, linseed can be used as raw, crushed, extruded or oil. Conjugated linoleic acid (CLA) isomers are a group of linoleic acid isomers, mainly contained in dairy products, that are considered to have anticarcinogenic, antiatherogenic, and antiobesity effects and immunomodulatory properties (McGuire and McGuire, 2000). Linseed is one of the vegetable fat sources with increased amount of C18:3n-3 (50% of the total FA). Feeding cows with extruded linseed increases milk unsaturated fatty acids (UFA) content; and in particular n-3 FA, *trans*-11 C18:1, and CLA concentrations, with reduced n-6 to n-3 fatty acid ratio (Fuentes *et al*., 2008). In particular, ruminant milk is rich in C18:2 *cis*-9,*trans*-11 CLA isomer, which is synthesized both in the rumen through the biohydrogenation of linoleic acid, and in the mammary gland through the desaturation of transvaccenic acid C18:1 *trans*-11 by Δ9-desaturase. The CLA content in cow milk can be manipulated by the diet. Vegetable oils rich in linolenic acid have been supplemented to dairy cows to enhance the milk content of C18:2 *cis*-9,*trans*-11 CLA (Zheng *et al*., 2005; Bu *et al*., 2007). Feeding flaxseed to dairy cows decreased the concentrations of short- and medium-chain FA and increased the long-chain FA content in milk fat (Mustafa *et al*., 2003; Petit, 2003). Therefore, the aim of the study was to evaluate the effects of two different lipid supplementations (crushed flaxseed *versus* flaxseed oil) on milk composition and milk FA profile of Damascus goats.

2. Materials and methods

2.1. Animals, feeding and experimental design

Fifteen lactating Damascus goats of 43.5±2 kg of live body weight (BW) were divided into 3 groups of 5 goats in a complete randomize design for 90 days. Goats were housed individually in tie stalls. A total mixed ration was formulated with 1:1 (DM) forage: concentrates ratio to meet their nutrient requirements as recommended in NRC (1981). The control diet

consisted of berseem clover (*Trifolium alexandrinum*) and concentrate mixture on a 1:1 DM basis. The other two diets contained control ration + 50g/head/day crushed linseeds (LS), and control ration +20 ml/head/day linseeds oil (LO). Diets were fed individually twice daily at 0800 and 1600 h in equal portions. The ingredient and nutrient composition of the diets are in Table (1). Dry matter intake was measured after 30, 60 and 90 days throughout the experimental period by weighing the offered diets and refusals from the previous day. Drinking water was available at all time. Individual milk samples were collected from all goats every two weeks during the experimental period. Goats were hand milked at 0900 and 2100 h, and portions according to milk yield of each milking were pooled for daily milk samples.

Table 1. Chemical composition (%; dry matter basis) of concentrate and berseem clover fed to lactating Damascus goats.

Items	Diet ingredients	
	CFM*	Berseem clover
Dry matter	91.29	13.3
Organic matter	89.89	88.2
Crude protein	14.15	14.2
Ether extract	4.05	2.6
Crude fiber	15.33	27.5
Nitrogen-free-extract	56.36	43.9

* The concentrates consisted of 25% undecorticated cotton seed meal, 35% wheat bran, 30% corn, 3% rice bran 3% molasses, 2% limestone, 1% urea and 1% salt (NaCl).

2.2. Ruminal fermentation

Rumen liquor samples were collected from three goats within each group by a stomach tube as described in Khattab *et al*. (2011). Collection was performed four hours after morning feeding, at 30, 60 and 90 days. The rumen samples were filtered through two layers of cloth, and used quickly as possible for the measurement of pH by using digital pH-meter. Strained rumen liquor was stored in glass bottles (25 ml) with few drops of toluene and paraffin oil just to cover the surface and stored at a deep freeze (-18°C) till chemical analysis.

2.3. Chemical analysis

Dried feed samples was ground through a Wiley mill (Arthur H. Thomas, Philadelphia, PA, USA) using a 1 mm screen. Samples of feed was analyzed for DM (#930.15), N (#954.01), and ether extract (EE; #920.39), according to AOAC (1995). Milk samples were analyzed using Foss 120 Milko-Scan, Foss Electric, Hillerød, Denmark. A digital pH meter with a glass electrode was used to determine ruminal pH, which were also analyzed for ammonia N by AOAC (1995). Ruminal total volatile fatty acids (VFA's) and fractions of volatile fatty acids were determined by Gas chromatography (Varian 3700; Varian Specialties Ltd, Brockville, Ontario, Canada). Milk fatty acids were determined using methyl esters prepared by base-catalyzed methanolysis of the glycerides (KOH in methanol) according to International Standards (ISO-IDF, 2002).

2.4. Statistical Analysis

Collected data on nutrient intake, ruminal fermentation parameters, milk yield and composition, and milk fatty acid profile were analyzed using the GLM procedure of SAS (SAS Inst. Inc. Cary, NC, 2006) in a complete randomized design. Significance was declared at a level of $P<0.05$ and trend of $P\leq0.10$ (Steel and Torrie, 1980).

3. Results

3.1. Feed intake and ruminal fermentation characteristics

Dry mater intake (DMI) values were not affected by LO or LS additives. Ruminal liquor pH was decreased ($P<0.05$) with LS and LO than control. However, total VFA and butyrate (C4) concentrations increased ($P<0.05$) with LS and LO. Acetate (C2) and propionate (C3) concentrations were higher ($P>0.05$) with experimental additives. Ammonia-N was lower ($P=0.08$) when goats fed LS or LO (Table 2).

3.2. Milk production and milk composition

Milk yield and fat content were higher ($P<0.05$) with goats fed LO followed by LS and then the control (Table 3). Protein content was increased ($P<0.05$) with LO compared to control, without significant differences between LS and control. Milk urea-N was lower ($P<0.05$) in LO and LS than in the control. Other milk components were not affected by additives (Table 3).

Generally, feed efficiency calculated as milk yield/DMI was significantly improved by LO and LS supplementation compared with control (Table 3).

3.3. Milk fatty acids composition

Goats fed on LO or LS had decreased ($P<0.05$) content of SFA and increased ($P>0.05$) the proportion of C18:3N3 milk fat. USFA were increased ($P<0.05$) with LO but; however, it increased ($P>0.05$) with LS compared to control. The proportions of conjugated linoleic acid (CLA) increased ($P<0.05$) with addition of LO or LS in the diet.

4. Discussion

4.1. Dry mater intake and rumen parameters

No effects were observed on DMI. The obtained results of DMI are in good agreement with a previous report by Benchaar *et al*. (2012), who studied the effect of increasing amounts of linseed oil to dairy cows.

A significant decreased in pH with LO and LS was observed. This may be a result of the higher in the VFA. Moreover, these additions is oily which may cause increased rumen acidity. Benchaar *et al*. (2012) reported no effect of linseed oil supplementation on ruminal liquor pH when they added LO at 3% to animal diets. The highest value ($P<0.05$) of the VFA's

were recorded with LO followed by LS and then control. These results suggest that the anaerobic fermentation of LO or LS treatments were more efficient and faster yielding more VFA than that in control by indirect way. These results can be supported by the data from Morsy *et al*. (2012), who observed an increased ruminal VFA in goats fed a concentrate-based diet and supplemented with different essential oils. However, Benchaar *et al*. (2012), found no effect of LO supplementation on VFA concentration. Supplementation with LO or LS resulted in increases molar proportions of butyrate and decreased acetate: propionate ratio. These results concur with those of Benchaar *et al*. (2012) in dairy cows. On the whole assumed that supplementation of unprotected and highly unsaturated fats to diets of ruminants decrease the acetate proportion and acetate: propionate ratio in the rumen, with an antimicrobial effect of PUFA rich oils as one probable explanation for this phenomenon (Jenkins and Jenny, 1992). Past studies reported a change in VFA patterns toward proportionately more propionate and less acetate when sheep (Broudiscou *et al*., 1994) and dairy cows (Ueda *et al*., 2003) were supplemented with LO. Lower ammonia-N concentration can be attributed to the action of LO and LS additives as regulators in absorbing and releasing ammonia within the rumen. These benefits might give favorable conditions in the rumen for microbial activity for better utilization of ruminal ammonia and useful conversion of N into microbial protein. These results may be due to the higher uptake of ammonia to microbial protein syntheses with animals fed LO or LS additives than with the control animals. Benchaar *et al*. (2012) found that no effect of LO supplementation on ammonia concentration. However, Ueda *et al*. (2003) observed increased ruminal ammonia concentration in LO supplemented dairy cows when compared with control cows. Other study reported a decrease in ammonia concentration in sheep supplemented with different levels of LO (Broudiscou *et al*., 1994).

4.2. Milk production and milk composition

This study showed higher milk production with goats fed LO or LS which were in the line of Gomez-Cortes *et al*. (2009) and Kholif *et al*. (2011) when used extruded linseed, and Benchaar *et al*. (2012) when used LO. These results are conflict with the findings of other studies, which found a reduction in milk production (Petit *et al*., 2005) or no response as a result of feeding whole flaxseed (Cortes *et al*., 2010), flaxseed oil (Caroprese *et al*., 2010) or extruded flaxseed (Lerch *et al*., 2012). The increased milk yield with treatments may be due to higher VFA concentration in rumen of goats given treated rations. Moreover, the higher milk fat may be due to the slightly increased ruminal acetate proportion (Palmquist and Beaulieu, 1993), in cows and (Gargouri *et al*., 2006) in sheep, who reported that an increase in the percentage of milk fat when used saturated or protected fats. Results of dietary supplementation with unprotected oils rich in PUFA are fully varied. With rapeseed oil, Mir *et al*. (1999) obtained a significant increased milk fat content in dairy goats. Milk protein was increased with additives as a result of improvement of ruminal microbial protein synthesis (Kholif *et al*., 2014). Similar results are obtained by Cortes *et al*. (2010), who found a higher milk protein with cows fed linseed oil.

4.3. Milk Fatty Acids

It is known that, fat source from the feed has an indirect effect on lipogenesis in the mammary gland, as it modifies ruminal fermentation. Reduction in SFA content and increment in USFA content in milk after dietary supplementation using LO or LS has been reported in numerous studies in cow (Ferlay *et al*., 2013), buffaloes (Kholif *et al*., 2011), and goats (Bernard *et al*., 2009). The increased proportions of C18:3N3 led to a decrease (P>0.05) in the ratio of n-6 to n-3 FA in milk fat of goats fed LO or LS. Lowering the ratio of n-6 to n-3 FA in food products has been recommended to prevent or modulate certain diseases in humans (Connor, 2000).

Table 2. Rumen fermentation parameters of lactating Damascus goats fed of diets supplemented with linseed or linseed oil (n=9).

Items	Diets[1]				
	Control	LS	LO	SEM	*P*-Value
Live body weight (kg)	44.70	44.98	44.74	0.319	0.917
Dry matter intake (kg/h/d)	1.36	1.34	1.38	0.017	0.081
Ruminal fermentation					
pH value	6.20[a]	6.03[b]	5.83[c]	0.057	0.002
VFA (mmol/L)	78.1[b]	87.1[a]	90.8[a]	1.97	0.00
Acetate (C2; mmol/100 mmol)	58.1	63.6	64.5	2.83	0.535
Propionate (C3; mmol/100 mmol)	28.8	32.5	33.5	1.41	0.414
Butyrate (C4; mmol/100 mmol)	10.2[b]	12.9[a]	14.1[a]	0.07	0.050
C2:C3	2.03	1.95	1.92	0.08	0.959
NH_3-N (mg/L)	283	261	265	5.79	0.086

[1]Control= concentrates and berseem clover (1:1), LS= control ration+50g/head/day crushed linseed, LO= control ration+20 ml/head/day linseed oil.
SEM= standard error of the means, VFA= volatile fatty acids.
Means at the same row with different superscript are significantly (P<0.05) different.

Table 3. Average daily milk yield (g/d) and composition (%) of lactating Damascus goats fed on diets supplemented with linseed or linseed oil (n=9).

Item	Diets[1]				
	Control	LS	LO	SEM	*P*-Value
Milk yield (g/d)	1140[b]	1230[a]	1256[a]	17.2	0.004
Milk composition (%)					
Fat	3.96[b]	4.27[a]	4.37[a]	0.054	0.001
Protein	3.16[b]	3.32[ab]	3.52[a]	0.067	0.050
Lactose	4.65	4.84	4.89	0.055	0.174
Total solids	12.8	13.6	13.5	0.196	0.191
Solids not fat	8.86	9.37	9.16	0.165	0.493
Ash	0.88	0.88	0.89	0.004	0.725
Urea N	39.2[a]	37.2[b]	36.4[b]	0.423	0.008
Feed efficiency					
Milk yield/DMI	0.83[b]	0.91[a]	0.91[a]	0.084	0.038

[1]Control= concentrates and berseem clover (1:1), LS= control ration+50g/head/day crushed linseed, LO= control ration+20 ml/head/day linseed oil
SEM= standard error of the means.
Means with different superscripts are significant (P<0.05) difference.

Table 4. Fatty acids (g/100 g total fatty acids) in milk of lactating Damascus goats fed on diets supplemented with linseed or linseed oil (n=9).

Item	Diets[1]				
	Control	LS	LO	SEM	*P*-Value
C4:0	1.60	1.50	1.20	0.091	0.185
C6:0	1.26[b]	1.50[ab]	1.64[a]	0.076	0.022
C8:0	2.62[a]	2.26[b]	2.21[b]	0.104	0.039
C10:0	6.63	5.35	4.91	0.242	0.338
C12:0	3.12	3.76	3.29	0.285	0.698
C14:0	9.93	9.55	9.93	0.332	0.762
C14:1	0.14[b]	0.18[b]	0.53[a]	0.072	0.000
C15:0	0.28	0.28	0.31	0.021	0.140
C16:0	28.3	28.1	25.2	1.12	0.718
C16:1	0.17[b]	0.38[b]	0.87[a]	0.015	0.000
C17:0	1.11[a]	0.39[b]	0.37[b]	0.124	0.000
C18:0	14.3	12.5	14.1	0.925	0.547
C18:1N9T	25.1	28.7	28.8	1.20	0.566
C18:1N9C	3.97[b]	3.94[b]	4.98[a]	0.250	0.050
C18:2 *trans*-10, *cis*-12	0.05[b]	0.08[a]	0.09[a]	0.007	0.030
C18:2 *cis*-9, *trans*-11	0.10	0.12	0.13	0.012	0.664
C18:3N3	0.10	0.15	0.15	0.011	0.202
C18:3N6	0.30	0.42	0.43	0.031	0.166
C20:0	0.85	0.85	0.86	0.098	0.988
SFA	70.0[a]	66.1[b]	64.0[b]	1.22	0.047
USFA	29.9[b]	34.0[ab]	36.0[a]	1.06	0.050
MUSFA	29.4	33.2	35.2	1.29	0.227
PUSFA	0.55	0.77	0.80	0.055	0.147
Total CLA	0.15[b]	0.20[a]	0.22[a]	0.011	0.007
N6/N3	3.00	2.80	2.86	0.173	0.736

[1]Control= concentrates and berseem clover (1:1), LS= control ration+50g/head/day crushed linseed, LO= control ration+20 ml/head/day linseed oil.
Means with different superscripts are significant (P<0.05) difference.
SFA=total saturated fatty acids, USFA= total unsaturated fatty acids, PUSFA=poly unsaturated fatty acids, CLA=conjugated linolenic acid, SEM= standard error of the means

Milk contents from CLA were increased with 46.6 and 33.3 % for LO and LS treatments, respectively compared with control. Food product from ruminants is a major dietary supply of CLA for humans and there is consensus that CLA is intermediates in the biohydrogenation of linoleic acid. The increased milk CLA and concentration with the LO or LS was expected. Total CLA are intermediates of the biohydrogenation of C18:2 *cis*-9 *cis*-12, which is present in high concentrations in linseed. Many authors indicated that the main proportion of CLA (from 64 to 98%) in milk fat is produced in the mammary gland by Δ^9-desaturase (Griinari *et al.*, 2000). The *cis*-9 *trans*-11 CLA content of milk fat in

the LO and LS diets came, in part, from ruminal biohydrogenation of linoleic acid and, in part, was produced by Δ^9-desaturase activity, from C18:1 *trans*-11. All *cis* and *trans*-isomeric combinations of CLA are known in food; however, C18:2 cis-9, trans-11 is the most common, although the presence of others CLA isomers and their probably concentrations vary according to rumen conditions (Kelly, 2001). There are other studies that noted increased CLA content in milk fat after supplementing diets with different linseed forms. These studies include those of Bu *et al*. (2007), in cow fed linseed oil, Bernard *et al*. (2009) in goats fed linseed oils, Gomez-Cortes *et al*. (2009), in ewes fed extruded linseed, Kholif *et al*. (2011) in buffaloes fed crushed flaxseed, and Lerch *et al*. (2012), Ferlay *et al*. (2013) in cow consuming extruded linseed.

5. Conclusion

Obtained results support the conclusion that addition of linseed oil to the ration of lactating goats, at the prescribed amount, was more effective in improving performance when compared with linseed. Adding linseed oil or linseed to the ration improved milk production, rumen activity and feed efficiency when compared with the control treatment.

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