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# Effect of whole or extruded linseed and of *Acacia cyanophylla* foliage intake in grazing dairy ewes

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**Abstract.** This experiment was conducted to investigate the effects of two different forms of linseeds (whole or extruded) and *Acacia Cyanophylla* tannins on milk yield and fatty acid (FA) profile and nitrogen balance in dairy ewes. Four groups of ten ewes, grazing triticale pasture, were supplemented with 250 g of soybean meal and 70 g of whole linseed (SW) or with 70 g of extruded linseed (SE), or with whole linseed and 100 g of acacia foliage (SWA) or with extruded linseed and 100g acacia foliage (SEA). Extruded linseed induced a significant decrease (P<0.05) of NDF digestibility of 2.2% over whole linseed. Intake of acacia foliage decreased (P<0.05) urinary nitrogen by 1.4 g/d and increased (P<0.0005) the nitrogen retention by 4.1 g/d. Milk production and composition were not affected by the diets except milk urea which decreased significantly (P<0.05) with extruded linseed and with the intake of acacia foliage. Stearic acid (C18:0) in ewe's milk was significantly decreased (P<0.006) with extruded linseed and with acacia and extruded linseed (P<0.03). Extruded linseed and acacia induced a decrease (P<0.0001) of oleic acid (C18:1 cis-9) by -1.82% and -1.23%. Acacia had no effects on omega-3 fatty acids and on the CLA, but fatty acid of the omega-6 group like $ -linolenic acid (C18:3 cis-6, cis-9, cis-12 (n-6)) were increased.

**Key words.** Linseed – Tannins – Nitrogen balance – Milk yield – Fatty acid – Ewe.

***Effet de la graine de lin entière ou extrudée et de l'ingestion de feuilles d'*Acacia cyanophylla *chez la brebis laitière au pâturage***

***Résumé.*** *Cette expérience a été réalisée pour étudier les effets de deux formes différentes de graines de lin (entier ou extrudé) et les tanins d'*Acacia cyanophylla *sur la production laitière, le profil des acides gras du lait et le bilan azoté chez les brebis laitières. Quatre groupes de dix brebis sur pâturage de triticale ont été complétés avec 250 g de tourteau de soja plus 70 g de lin entier pour le lot (SW) et 70 g de lin extrudé pour le lot (SE). Les brebis des lots (SWA) et (SEA) reçoivent en plus par rapport aux lots (SW et SE) 100 g de feuillage d'acacia. Le lin extrudé a induit une diminution significative (P<0,05) de la digestibilité du NDF de 2,2% par rapport au lin entier. L'ingestion d'acacia a diminué (P<0,05) l'azote urinaire de 1,4 g/j et a augmenté (P<0,0005) la rétention d'azote de 4,1 g/j. La production et la composition du lait n'ont pas été affectées par les régimes à l'exception de l'urée du lait qui a diminué de façon significative (P<0,05) avec l'ingestion de graines de lin extrudées et d'acacia. L'acide stéarique (C18:0) dans le lait des brebis a diminué de manière significative (P<0,006) avec l'ingestion du lin extrudé seul et avec l'ingestion d'acacia et de lin extrudé (P<0,03). Les graines de lin extrudées et l'acacia ont induit une baisse (P<0, 0001) de l'acide oléique (C18:1 cis-9) de -1,82% et -1,23%. L'acacia n'a eu aucun effet sur les acides gras du groupe oméga-3 et sur le CLA, mais les acides gras du groupe oméga-6 comme l'acide * -linolénique (C18:3 cis-6, cis-9, cis-12 (n-6)) a augmenté.*

***Mots-clés.*** *Lin – Tanins – Bilan azoté – Production laitière – Acide gras – Brebis.*

## I – Introduction

Oilseeds could be incorporated in ruminant diets in order to increase polyunsaturated fatty acids (PUFA), content in dairy products, and thus to improve their nutritional quality (Shingfield *et al*., 2013). The changes in PUFA content of dairy products depend on the basal diets and oilseeds. In manufacturing feed, the extrusion technique is used to destroy the enzymes which are responsible for rancidity and anti-factors' nutrient, present in the seeds. This reduces hydrogenation of unsaturated FA in the rumen. *Acacia Cyanophylla* contains tannins, which could partially protect dietary proteins and lipids from rumen metabolism by modifying environment in rumen and microorganisms' activity. The objective of this experiment was to study the impact of the form of linseed (whole or extruded) and of a supplementation with acacia foliage on sheep milk production fatty acid profile and nitrogen balance.

## II – Materiel and methods

The experiment was carried out in the dairy experimental farm of the National Institute of Agricultural Research of Tunisia (INRAT), on 40 Sicilo-Sarde breed ewes divided into four homogeneous groups: 10 ewes each, according to their age, parity and milk production (567 ml/day). Ewes were conducted together during the day (from 1000 h to 1500 h) on triticale pasture with rotational grazing system with a stocking rate of 26 ewes/ha. Indoors, ewes were separated and lodged per groups, receiving 250 g of soya bean meal. Such diet was complemented by 70 g of whole linseed for the first group (SW), 70 g of extruded linseed for the second group (SE), 70 g of whole linseed together with 100 g *Acacia Cyanophylla* foliage for the third group (SWA) and 70 g of extruded linseed with 100 g acacia foliage for the fourth group (SEA). The experiment lasted 70 days. Five animals from each group were placed in individual metabolic cages during 2 weeks for a digestion trial (10-day adaptation period, 5-day period of total faecal and urine collection). Concentrate, linseed and acacia were offered once daily at 09:00. Fresh cut triticale and oat silage were distributed in two equal meals at 09:00 and 16:00 in separate troughs. Milk fat, protein, and urea N were analyzed by a MilkoScan 4000 (Foss Electric, integrated Milk Testing). Methyl esters of FA were analyzed by gas chromatography (GC) (Shimadzu, GC-14 A). Data of milk yield, composition, fatty acid profile, diet digestibility and nitrogen balance (twice a month for two months and a half), were analyzed as repeated measures using the mixed procedure of SAS (2000). The statistical model included: experimental diet, time of sampling and their interactions as fixed effects. The data recorded during pre-experimental period (milk yield and composition) were used as covariates and included in the model. The following contrasts were used to compare the effects of different diets: Contrast 1: linseed form effect {SW, SWA vs. SE, SEA}; Contrast 2: acacia effect {SW, SE vs. SWA, SEA}; Contrast 3: acacia effect according to linseed form {SWA vs. SEA}.

## III – Results and discussion

The chemical composition of experimental feeds was presented in Table 1. The protein content of triticale was around 9.7%. This content is considered low as a consequence of the absence of nitrogen fertilization during growing period.

The feed intake was average of 1.84 kg/ewe/d and the total intake of 2.17 kg/ewe/d. The digestibility of organic matter did not varied with diets and reached 83.8% (Table 2). The digestibility of CP tended to differ ($P < 0.10$) between groups. According to contrasts studied, extruded linseed tended ($P = 0.08$) to increase the CP digestibility by 1.9% over the whole linseed (contrast C1). *Acacia cyanophylla* (contrast C2) tended ($P = 0.09$) to increase the CP digestibility by 1.9%. The same for the contrast C3, the contribution of acacia with extruded linseed tended ($P = 0.08$) to increase the CP digestibility by 2.6%. These results could be explained partly by the fact that the extrusion reduces ruminal degradability of protein in rumen. The digestibility of the NDF fraction was significantly decreased by 2.2% with extruded linseed compared to whole linseed.

**Table 1. Chemical composition of triticale, Soya bean meal, linseeds (whole and extruded) and *Acacia Cyanophylla* (in% of DM)**

| Chemical composition | Triticale | Soya bean meal | Whole linseed | Extruded linseed | *Acacia cyanophylla* |
|---|---|---|---|---|---|
| Dry matter (DM) (%) | 23.7 | 85.4 | 92.2 | 92.5 | 97.7 |
| Organic matter | 93.5 | 93.2 | 96.3 | 95.5 | 90.8 |
| Crude protein | 9.7 | 42.9 | 19.6 | 19.2 | 14.3 |
| Neutral-detergent fibre | 53.9 | 34.4 | 56.2 | 48.6 | 44.5 |
| Acid-detergent fibre | 29.9 | 6.6 | 33.7 | 27 | 38.2 |
| Fat matter | – | – | 29 | 30.8 | – |
| Ash | 6.5 | 6.8 | 3.7 | 4.5 | 9.2 |
| Total phenols† | – | – | – | – | 27.5 |
| Condensed tannins†† | – | – | – | – | 32.7 |
| Palmitic acid | – | – | 6.6 | – | – |
| Stearic acid | – | – | 4.13 | 4.2 | – |
| Oleic acid | – | – | 18.4 | 18.6 | – |
| Linoleic acid | – | – | 14.1 | 14.3 | – |
| Linolenic acid | – | – | 53.8 | 53.1 | – |

† Expressed as g tannic acid equivalent per kg DM. †† Expressed as g leucocyanidin equivalent per kg DM.

**Table 2. Effect of experimental diets on forage and dry matter intake and diet digestibility in dairy ewes**

| | SW | SE | SWA | SEA | m.s.e | Pr.>F | Contrasts Pr.> F | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | C1 | C2 | C3 |
| DM forage intake (g/d) | 1742 | 1866 | 1892 | 1892 | 156.7 | 0.5 | 0.4 | 0.3 | 0.9 |
| Total DM intake (g/d) | 2020 | 2144 | 2267 | 2267 | 156.7 | 0.1 | 0.4 | 0.04 | 0.9 |
| OM digestibility (%) | 83.6 | 83.2 | 83.5 | 85 | 1.86 | 0.4 | 0.4 | 0.3 | 0.2 |
| CP digestibility (%) | 79.5 | 80.6 | 80.6 | 83.2 | 2.02 | 0.1 | 0.08 | 0.09 | 0.08 |
| NDF digestibility (%) | 81.8 | 78.4 | 82.5 | 81.5 | 2 | 0.06 | 0.03 | 0.1 | 0.3 |

m.s.e: mean standard error.

Acacia supply increased ($P < 0.0001$) the nitrogen intake (Table 3). Diets did not significantly affect faecal nitrogen (g/day), but C1 and C3 contrasts showed significant differences. In fact, extruded linseed intake and with *Acacia cyanophylla* induced a decrease of faecal nitrogen (0.5 g/day and 0.8 g/day respectively) compared with whole linseed intake.

**Table 3. Effect of experimental diets on nitrogen balance in dairy ewes**

| | SW | SE | SWA | SEA | m.s.e | Pr.>F | Contrasts Pr.> F | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | C1 | C2 | C3 |
| N intake (g/day) | 27.4 | 27.9 | 30.4 | 30.3 | 0.8 | 0.0002 | 0.5 | 0.0001 | 0.9 |
| Fecal N (g/day) | 5.6 | 5.4 | 5.9 | 5.1 | 0.5 | 0.1 | 0.05 | 0.9 | 0.03 |
| Urinary N (g/day) | 9.5 | 9.8 | 8.4 | 8.1 | 1.2 | 0.2 | 0.9 | 0.04 | 0.7 |
| N retention (g/day) | 12.2 | 12.8 | 16.1 | 17.1 | 1.6 | 0.002 | 0.3 | 0.0002 | 0.3 |
| N Absorbed (g/day) | 21.7 | 22.5 | 24.5 | 25.2 | 1.2 | 0.002 | 0.2 | 0.0004 | 0.3 |

m.s.e: mean standard error.

Urinary nitrogen (g/day) ranged from 8.1 g/day and 9.8 g/day, without significant effects of diets. Acacia (contrast C2) induced a significant decrease (P< 0.05) in urinary nitrogen by 1.4 g/day and a significant increase in nitrogen retention (g/day) of 4.1 g/day. For the absorbed nitrogen (Nab/g/day), the respective values ranged from 21.7 g/day to 25.2 g/day, with significant differences (P<0.003) between diets. This parameter increased significantly with acacia supply (C2, P<0.0005) by 2.8 g/day.

Milk production as well as fat and protein contents were not significantly affected by the diets (Table 4). Fat and protein content were on average 7.3 and 4.8%, respectively. Milk urea, significantly (P<0.05) varied according to experimental diets. Indeed, extrusion of linseeds notably (P=0.05, contrast C1) decreased milk urea by about 3 mg/dl. Similarly, *Acacia Cyanophylla* supply (contrast C2) induced a significant (P<0.05) decrease of milk urea by 3.8 mg/dl, which could be a result of a better use of food proteins, following their protection by tannins in the rumen. Milk urea for ewes fed with extruded linseeds and tannins of acacia has the lowest value. This result agrees with that of Maamouri *et al.* (2011), who suggested that *Acacia Cyanophylla* supply much decreased ewe's milk urea. The application of heat (extrusion) may reduce the bacterial protein de-amination in the rumen. Stearic acid (18:0) varied significantly (P <0.04) between groups. Indeed, this fatty acid considerably decreased (contrast C1, P <0.006) with extruded linseed intake (-2.5%). The decrease was also about -2.2% with *Acacia Cyanophylla* (C3 contrast, P <0.03). Oleic acid (c9-18:1), the second most important fatty acid in milk' ewes, with an average percentage of 18.8%, was significantly decreased by extruded linseed and Acacia Cyanophylla (P<0.0001). The percentage (% of total FA) of rumenic acid (c9, t11-18:2, CLA isomer) (P<0.05) increased by 0.5% in the sheep milk with extruded linseed (C1, P<0.05). *Acacia Cyanophylla* also increased by 0.7% the content of this particular fatty acid in milk with extruded linseed (C3, P<0.05). Finally *Acacia Cyanophylla* intake induced an important increase (C2, P<0.05) for "n-6" FA such as $ -linolenic acid (c6, c9, c12-18:3 (n-6)).

**Table 4. Effect of experimental diets on milk yield and fatty acid composition (g/100g of Tot FA) in dairy ewes**

| | SW | SE | SWA | SEA | m.s.e | Pr.>F | Contrasts Pr.> F | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | C1 | C2 | C3 |
| Milk yield (ml/day) | 630 | 641 | 628 | 692 | 5.6 | 0.6 | 0.3 | 0.5 | 0.3 |
| Fat content (%) | 7.4 | 7.4 | 7.4 | 7.2 | 0.05 | 0.8 | 0.7 | 0.6 | 0.5 |
| Protein content (%) | 4.9 | 4.9 | 4.8 | 4.8 | 0.03 | 0.7 | 0.7 | 0.3 | 0.9 |
| Milk urea N (mg/dl) | 27.8 | 24.1 | 23.7 | 20.6 | 0.8 | 0.05 | 0.05 | 0.03 | 0.2 |
| C18:0 | 13.5 | 10.7 | 12.7 | 10.5 | 0.3 | 0.04 | 0.006 | 0.4 | 0.03 |
| c9-18:1 | 20.9 | 17.8 | 18.4 | 17.9 | 0.2 | 0.0001 | 0.0001 | 0.0001 | 0.0002 |
| c9, t11-18:2 | 1.03 | 1.3 | 0.88 | 1.6 | 0.06 | 0.05 | 0.01 | 0.7 | 0.01 |
| c9, c12, c15-18:3 (n-3) | 1.06 | 1.3 | 0.99 | 1.27 | 0.03 | 0.14 | 0.03 | 0.7 | 0.08 |
| c6, c9, c12-18:3 (n-6) | 0.09 | 0.11 | 0.12 | 0.13 | 0.004 | 0.13 | 0.19 | 0.04 | 0.4 |

m.s.e: mean standard error.

## IV – Conclusion

Extruded linseed decreased NDF digestibility. Acacia intake seems to play a role in the decrease of urinary nitrogen and an increase of the nitrogen retention while did not affect milk production and composition except milk urea which also decreased with extruded linseed intake in comparison with whole linseed. On the fatty acid profile of milk, extruded linseed and acacia induced a decrease of oleic acid (C18:1 cis-9). The extrusion of linseed leads to an increase in fatty acids omega-3 group and the CLA.

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