

Article

Obtaining Goats' Dairy Products Enriched in Healthy Fatty Acids by Valuing Linseed or Hempseed as Dietary Ingredients

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Abstract: The study aimed to assess the effects of including linseeds or hempseeds in the diets of late lactation Murciano-Granadina dairy goats on the nutritional quality of the milk and cheese fat, expressed by the fatty acids profile and the healthy lipid indices. Thirty-six goats were randomly distributed in 3 groups of 12 animals each, according to a 3 × 3 Latin square design, and fed three different diets: group CON (control, with sunflower meal, 11.5% DM basis); group LIN, where sunflower meal was replaced by linseed; and group HMP, where sunflower meal was replaced by hempseeds. The replacement had no effects on the milk yields and the milk protein content as no significant differences were detected among groups. The significant increase of the fat content in the case of the LIN and HMP groups was accompanied by significant decreases in saturated fatty acids concentration and very significant increases in monounsaturated fatty acids. The content of n3 and n6-PUFAs (polyunsaturated fatty acids) increased, mainly due to a 4.1 times higher proportion of alpha-linolenic acid (ALA; C 18:3n-3) in LIN diet milk and a 1.3 times higher proportion of linoleic acid (LA; C 18:2n6c) in HMP diet milk. The conjugated linoleic acid (CLA; isomer c9, t11) was 1.9 times higher for the LIN diet and 5.05 times higher for the HMP diet. Feeding either linseed or hempseeds contributed to the reduction of the atherogenic and thrombogenic indices, increased the hypocholesterolemic: hypercholesterolemic ratio as well as the proportion of other desired fatty acids in the milk fat. The improved nutritional quality of milk, which has potentially far-reaching human health benefits, is maintained in cheese through the increase of the n3 and n6-PUFAs, especially for the LIN diet where the n6/n3 ratio decreased significantly, compared with the CON diet (3.62 vs. 6.88). The CLA concentration was significantly higher ($p < 0.001$) for the HMP cheese compared with the CON diet (1.89% vs. 0.78%). These effects highlight the opportunity of obtaining dairy products with improved nutritional quality using local feed resources.

Keywords: CLA; fatty acids; goat cheese; hempseed; linseed; milk



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1. Introduction

The existing knowledge regarding the potential effects of fats on human health allows food to be classified as “good” or “bad”, depending on the nature of the fats they contain. From a human health perspective, the fat in the human diet should have an ideal fatty acids (FA) composition of 8% SFA (saturated fatty acids), 82% MUFA (monounsaturated fatty acids), and 10% PUFA (polyunsaturated fatty acids) [1]. Dairy products are a good source of fat in human nutrition but at the same time, dairy fat contains, on average, 70% SFA,

25% MUFA, and 5% PUFA and therefore may induce health problems like cardiovascular diseases. But the profile of FA can be healthier, especially by tailoring the feeding strategies for ruminants [2] to produce a shift of the of n6/n3 PUFAs ratio in the dairy products [3] or an improvement of the CLA content [4].

Goat milk and goats' dairy products are already associated by consumers with the image of healthy food; this offers the premises for introducing more valuable goat milk products to the market, by feeding the goats with dietary ingredients that are rich in PUFA, in quantities that allow detectable effects in milk. Such ingredients, rich in n3 or/and n6-PUFA, are oilseeds [5]; however, for some of them or their by-products, the effects on goats are less studied. For example, their high fat content may be a limitation of their use in goats' diets, as it may impair rumen processes such as cellulolysis. On the other hand, they should be included in proportions that are high enough to overcome rumen biohydrogenation of the unsaturated fatty acids.

The linseeds (*Linum usitatissimum* L.) contain oil with a high proportion (more than 50%) of ALA (alpha-linolenic acid; C 18:3n3), an important n3-PUFA acid, and they have been used successfully in the diets of ewes to produce n-3 enriched milk as mentioned by some authors [6–8]. More recently, Rapetti et al. [9] reported that after feeding linseeds and hempseeds (at a 9.3% proportion in the diet total DM), Alpine goat milk had higher levels of ALA; linseed induced the lowest n-6/n-3 ratio of the experimental groups, while the in case of LA, no differences were registered among diets.

The hempseeds (*Cannabis sativa* L.) were reconsidered for the use in dairy ruminant nutrition because of their high level of valuable PUFA–LA (linoleic acid; C 18:2n6) as the most predominant FA (53.4–60.0%) [10], followed by ALA (12.98–22.40%) [11], oleic, palmitic, and stearic acids. There are some examples of its inclusion in the nutrition of dairy small ruminants. The hempseeds were utilized successfully to manipulate the FA profile in the sheep milk [12], as well as in Carpathian goat milk with a diet with 4.7% hempseed oil on DM [13] or 9% hempseeds on DM [14]. Also, Cremonesi et al. included hempseeds in diets for Alpine lactating goats by 9.3% on DM [15] or 9.4% on DM [10], and their influence in the milk FA was noticeable. Also, an increased content of milk n3-PUFA and an improved n-6/n-3 ratio were obtained by Mierlita et al. [16] on Turcana dairy sheep after feeding with hempseeds and cake. Total CLA content increased by 2.0 times in the milk of the ewes that received hempseed and by 2.4 times with the hemp cake inclusion. The milk yield and milk fat content were increased but milk lactose decreased.

In this context, we intended to obtain enriched n3 and n6-PUFA milk from Murciano-Granadina goats, adapted to be raised in a farm from Romania using local linseeds or hempseeds in their diets. Therefore, the study aimed to assess the effects of the inclusion of 11.5% of linseeds or hempseeds (DM basis) in the hay-based diets on milk production, as well as on the fatty acid profiles of both milk and cheese.

2. Materials and Methods

2.1. Animals and Experiment Design

The study was carried out on a commercial farm in the southeast of Romania on Murciano-Granadina multiparous goats in late lactation, which have similar age, milk yield, and days in milk (Table 1). Thirty-six goats were randomly distributed in 3 groups of 12 animals each. According to a 3 × 3 Latin square design, each of the three groups was fed, in three successive periods, 3 experimental diets: CON (control group, based on sunflower meal), LIN (where sunflower meal was replaced by linseed), and HMP (where sunflower meal was replaced by hempseed) (see the Scheme 1 below). Each period of feeding lasted 28 days, of which 21 days were for adaptation to the diets and 7 days were for data recording and samplings.

The animals were kept indoors during the experiment, with free access to paddocks, and were group-fed. The animals were milked at 7:00 a.m. and 4:30 p.m., in a milking parlor allowing record of individual milk yields.

Table 1. Parameters of the goats' groups of at the beginning of the experiment.

	CON	LIN	HMP
Age, years	3.30	3.28	3.32
Days in milk	143.75	139.75	141.33
Average milk yield, kg/day	1.10	1.11	1.17

CON, control diet; LIN, linseed diet; HMP, hempseed diet.

	Group 1	Group 2	Group 3
Period 1	CON	LIN	HMP
Period 2	HMP	CON	LIN
Period 3	LIN	HMP	CON

CON, control diet; LIN, linseed diet; HMP, hempseed diet; Period 1-3, the three successive periods of feeding.

Scheme 1. The scheme of 3×3 Latin square experiment design.

2.2. Diets and Ingredients

The diets were formulated according to the French feeding system [17]; the control group was fed a diet that is typical for the indoor feeding of goats in Southeastern Europe: hay and a mixture of cereals and sunflower meal (3:1). In the experimental diets, the sunflower was totally replaced with linseed or hempseed in order to supply high amounts of PUFA-rich lipids. For practical reasons, no adjustments of the other dietary ingredients were made; therefore, the experimental diets had a higher energy supply and slightly lower protein supply. The goats were group-fed by limited amounts of compound feed (1.2 kg/head/day) and hay (1.5 kg/head/day) as presented in Table 2. The ingredients of the compound feeds were grinded including the oilseeds. Diets were fed twice daily in equal amounts (at 8.00 a.m. and 5.00 p.m.). Access to water and a trace-mineralized salt block was provided *ad libitum*.

Table 2. Consumption of diets and daily nutritive supplies.

Ingredients	CON	LIN	HMP
Diets' consumption, kg/day			
Grass hay	1.3	1.3	1.3
Alfalfa hay	0.2	0.2	0.2
Compound feed	1.2	1.2	1.2
Compound feeds' structure, %			
Maize (%)	33.4	33.4	33.4
Barley (%)	8.3	8.3	8.3
Oat (%)	12.5	12.5	12.5
Wheat (%)	8.3	8.3	8.3
Wheat bran (%)	8.3	8.3	8.3
Sunflower meal (%)	25.0	0.0	0.0
Linseeds (%)	0.0	25.0	0.0
Hempseeds (%)	0.0	0.0	25.0
Calcium carbonate (%)	2.2	2.2	2.2
Salt (%)	1.0	1.0	1.0
Vitamin-mineral premix for goat (%)	1.0	1.0	1.0
Nutritional and fatty acids supply from the consumed diets			
Dry matter, kg/day	2.356	2.356	2.358
MFU(UFL)/kg DM/day	0.787	0.865	0.863
PDIN, g/day	219.05	181.90	185.86

Table 2. *Cont.*

Ingredients	CON	LIN	HMP
PDIE, g/day	192.67	183.48	190.01
Ether extract, g/day	30.70	107.85	109.29
Calcium, g/day	20.74	20.93	20.10
Phosphorus, g/day	12.03	9.70	9.51
C 16:0, g/day	5.61	9.71	10.62
C 18:0, g/day	0.89	3.61	2.82
C 18:1, g/day	6.32	20.72	15.49
C 18:2n-6, g/day	10.11	21.23	48.52
C 18:3n-3, g/day	2.45	39.08	15.64
Others, g/day	1.27	2.06	4.98
Total FA, g/day	26.66	96.41	98.06

CON = control diet; LIN = linseed diet; HMP = hempseed diet; MFU = Milk Feed Units (UFL), according to INRA system, 2010; 1 UFL = 1700 kcal; PDIN = protein truly digested in the small intestine when the protein is the limiting factor, according to INRA system, 2010; PDIE = protein truly digested in the small intestine when the energy is the limiting factor, according to INRA system, 2010.

The proximate composition of every dietary ingredient was assessed using commonly accepted methods [18]: dry matter (DM) by the gravimetric method, crude protein (CP) by the Kjeldahl method, crude fiber (CF) by successive hydrolysis in alkali and acid environment, ether extractives (EE) by extraction in organic solvents, and ash determined by the gravimetric method.

The levels of CP and CF for both oilseeds were comparable (Table 3). As for the composition of fatty acids, the Romanian variety of linseeds used in the experiment contained 51.33% ALA, 20.83% oleic acid, and 16.42% LA. The hempseeds utilized in the experiment were also sourced from a Romanian variety, primarily cultivated for oil production and characterized by low concentrations of delta-9-tetrahydrocannabinol. The hempseeds contain a high concentration (over 80% of the oil) of long-chain essential PUFAs, as detailed in Table 3. Specifically, they consisted of 53.41% LA and 18.08% ALA. Oleic acid content was 13.19% whereas palmitic and stearic acids were also present.

Table 3. Chemical composition and summarized fatty acids profile of the local varieties of linseeds and hempseeds and of the sunflower meal.

	Linseeds	Hempseeds	Sunflower Meal
Chemical composition			
Dry matter, g/kg	904.06	910.62	902.88
Crude protein, g/kg DM	228.35	236.36	450.99
Crude fat, g/kg DM	291.12	293.60	6.67
Crude fibre, g/kg DM	290.43	303.11	158.88
Nitrogen-free extract, g/kg DM	148.77	126.54	298.98
Ash, g/kg DM	41.32	40.38	84.47
Fatty acids (g/100 g total fatty acids)			
C 16:0	6.09	7.19	14.97
C 18:0	4.00	2.82	7.72
C 18:1	20.83	13.19	28.57
C 18:2n-6	16.42	53.41	37.04
C 18:3n-3	51.33	18.08	0.58
Others	1.44	6.12	2.07

C 16:0, palmitic acid; C 18:0, stearic acid; C 18:1, oleic acid; C 18:2n-6, linoleic acid; C 18:3n-3, alpha-linolenic acid.

2.3. Milk Samples

Two sets of milk samples were collected from each animal at the end of the experimental periods. The first set of samples was used for proximate analyses and therefore preserved with bronopol and stored at 4 °C until further analysis by infra-red spectroscopy.

The second set of milk samples was frozen (-18°C), without preservatives, for future assessment of the fatty acids' profile.

The proximate milk analysis for fat, protein, lactose, casein, urea, specific density, pH, and total solids was done by FTIR (Fourier Transform Infrared Spectrometer) rotation scanning with a CombiScope FTIR 200 device (Delta Instruments, Drachten, Holland) (ISO 9622:2013) [19]. The somatic cell count was determined according to the SR EN ISO 13366-2:2007 method [20].

The milk fatty acids composition was determined by the gas chromatography method after extracting the lipids by the EN ISO 661:2005/AC:2006 [21] method and then trans-methylating the fatty acids with a mixture of concentrated H_2SO_4 (95%) and methanol. The resulting methyl esters of fatty acids (FAME) were separated using the gas chromatograph GC Perkin Elmer-Clarus 500, equipped with a capillary column of high-polarity stationary phase (Agilent BPX70; $60\text{ m} \times 0.25\text{ mm}$ inner diameter $\times 0.25\text{ }\mu\text{m}$ thick film) and flame-ionization detector according to SR CEN ISO/TS 17764-2:2008 [22].

2.4. Cheese Samples

The cheese was manufactured from the milk distinctly collected from each experimental group. Ten pieces of cheese were manufactured for each group using a method that is common in the region. The raw milk, not standardized for fat content, was filtered, heated at 36°C , treated with Rennet 8 g (Ideal Still Exim SRL, Chitila, Romania) as a coagulant, thoroughly mixed, stabilized, and left for 60 min to coagulate. After the milk had clotted, the curd was cut with a knife and left to express the whey for 15 min. After the elimination of the whey, the cheese mass was placed in cheese cube molds. The cheese cubes were pressed for 1.5 h at 20°C , then cooled (6°C). After being brined in a solution containing 18% NaCl for 16 h at a temperature of 10°C , the cheese cubes were subsequently dried. They were then stored in a refrigerated environment at 5°C until further assays were conducted.

The DM and the CF content (g/100 g) for each of the 30 manufactured cheese pieces were determined according to the same methods that were used for diet ingredients. The fatty acid profile was determined following fat extraction from dried cheese, conversion to fatty acid methyl esters (FAME), and GC analysis performed according to the method described for the milk samples.

The FAME peak areas were converted to fatty acid (FA) using the FAME-to-FA conversion factor for milk. FAs were expressed as a percentage of the total identified FA (% of total FA), or in gravimetric contents (mg/100 g cheese), using the conversion factor for milk and milk products (0.945) for the calculation of total FA from total lipids [23].

2.5. Health Lipid Indices of Milk and Cheese Fat

In addition to the profile of individual FA with each diet, the proportion of beneficial FA was also calculated using the following parameters: PUFA/SFA ratio values; proportion of desired fatty acids (DFA); HSF (hypercholesterolemic SFA) and hypocholesterolemic/hypercholesterolemic (h/H) ratio. The nutritional quality of milk and cheese fat was also assessed by the calculation of health indices: the atherogenic index (AI), calculated according to [24], and the thrombogenic index (TI), calculated following [25].

2.6. Statistical Analysis

The effects of dietary inclusion of linseed or hempseed on productive performances, milk and cheese quality, and health indices were analyzed using the general linear model procedure (Minitab version 17, SAS Institute Inc., Cary, NC, USA), according to the following model:

$$Y_{ijk} = \mu + A_i + B_j + C_k + e_{ijk}$$

where Y represents variable studied during the trial; μ represents the overall mean; A represents the effect due to the treatment/diet (CON, LIN and HMP); B represents the effect due to the period (blocking factor); C represents the effect due to the group (blocking factor); and e represents the error.

Significance was declared at $p < 0.05$, while values between 0.05 and 0.1 were considered as tendencies.

3. Results

3.1. Proximate Composition of Milk and Cheese

No significant differences of the milk yield were detected among the three groups (1.18 L/head/day for CON, 1.14 L for LIN and 1.19 L for HMP, $p = 0.686$). Consequently, as the diets were fed in limited amounts (1.2 kg compound feeds/head, 1.5 kg hay/head), the feed consumption: milk yield ratio was not significantly changed by the inclusion of either linseeds or hempseeds: 2.06 kg DM/L of milk (CON), 2.26 kg DM/L of milk (LIN) and 2.08 kg DM/L of milk (HMP).

Also, in the case of protein and casein content, there were no significant differences among groups (Table 4).

Table 4. The milk yield and the proximate composition of the milk.

Specification	CON	LIN	HMP	SEM	p-Value		
					Treatment	Period	Group
Milk yield, kg/day	1.18	1.14	1.19	0.043	0.686	0.190	0.063
Milk fat, %	5.58 ^b	6.18 ^a	6.10 ^a	0.113	0.0001	0.805	0.0001
Milk protein, %	4.03	3.99	3.96	0.032	0.570	0.0001	0.749
Lactose, %	4.75 ^b	4.83 ^a	4.83 ^a	0.017	0.001	0.0001	0.012
Casein, g/L	32.99	32.72	31.89	0.443	0.185	0.0001	0.563
Urea N, mg/100 mL	48.49 ^a	40.14 ^b	41.98 ^b	1.323	0.0001	0.0001	0.050
Specific density, g/L	1026.87	1026.84	1026.83	0.103	0.951	0.005	0.0001
pH	6.57	6.57	6.58	0.017	0.850	0.0001	0.037
Total solids, %	15.27 ^b	15.78 ^a	15.69 ^{ab}	0.150	0.040	0.002	0.001
Somatic cells count × 1000 per mL	1943	2403	2354	235	0.312	0.119	0.001

CON, control diet; LIN, linseed diet; HMP, hempseed diet; SEM, standard error of the mean; ^a, ^b—means in rows marked with different uppercase superscripts significantly differ at $p < 0.001$; $n = 36$, number of milk samples analyzed.

On the other hand, milk fat content was significantly higher ($p = 0.0001$) in the case of the LIN (6.18%) and HMP (6.10%) diets compared to the CON diet (5.58%). Also, milk lactose content was significantly higher ($p = 0.001$) in the case of the LIN and HMP diets compared to the CON diet. A highly significant ($p = 0.0001$) decrease in the urea-N percentage was recorded with the LIN and HMP diets. This parameter is a predictor of nitrogen excretion and is linearly correlated with dietary crude protein content.

The other parameters for milk, like pH, density, and the number of somatic cells, were not influenced by the diets, with similar values being recorded among the three experimental groups. Only total solids for the LIN diet slightly differed from CON diet ($p = 0.04$).

The fat and protein contents of the goat cheese were also assessed, as shown in Table 5, and no significant differences were observed among the diets.

Table 5. The proximate composition of the cheese.

Specification	CON	LIN	HMP	SEM	p-Value
Goat cheese fat, %	21.67	21.26	22.03	1.00	0.864
Goat cheese protein, %	15.32	14.28	14.10	0.55	0.258

CON, control diet; LIN, linseed diet; HMP, hempseed diet; SEM, standard error of the mean; $n = 30$, number of cheese samples analyzed.

3.2. Profile of Fatty Acids in Milk

The particular oil composition of the local varieties of linseeds and hemp is reflected by the results of analyses of the milk fatty acids profile, presented in Table 6.

Table 6. The effect of linseeds and hempseeds in goats' diets on fatty acids milk composition (g FAME/100 g total FAME).

Fatty Acids in Milk		CON	LIN	HMP	SEM	p-Value		
						Treatment	Period	Group
Butyric	C 4:0	0.04 ^b	0.05 ^b	0.06 ^a	0.006	0.013	0.194	0.897
Caproic	C 6:0	1.32	1.31	1.42	0.038	0.057	0.124	0.597
Caprylic	C 8:0	3.11 ^{ab}	3.07 ^b	3.20 ^a	0.041	0.044	0.147	0.0001
Capric	C 10:0	12.09 ^a	10.67 ^c	11.12 ^b	0.143	0.0001	0.006	0.0001
Undecanoic	C 11:0	0.44 ^a	0.35 ^b	0.38 ^b	0.013	0.0001	0.987	0.185
Lauric	C 12:0	7.47 ^a	5.56 ^b	5.71 ^b	0.167	0.0001	0.0001	0.459
Tridecanoic	C 13:0	0.17	0.15	0.15	0.008	0.250	0.700	0.640
Myristic	C 14:0	11.98 ^a	9.46 ^b	9.64 ^b	0.175	0.0001	0.010	0.953
Miristoleic	C 14:1	0.78 ^a	0.57 ^b	0.59 ^b	0.022	0.0001	0.002	0.025
Pentadecanoic	C 15:0	0.31 ^a	0.28 ^b	0.26 ^b	0.008	0.001	0.0001	0.179
Pentadecenoic	C 15:1	1.05 ^a	0.96 ^b	0.96 ^b	0.021	0.002	0.0001	0.001
Palmitic	C 16:0	26.23 ^a	21.31 ^c	22.64 ^b	0.360	0.0001	0.004	0.802
Palmitoleic	C 16:1	1.98 ^b	1.78 ^c	2.23 ^a	0.042	0.0001	0.366	0.328
Heptadecanoic	C 17:0	0.36	0.35	0.34	0.008	0.372	0.001	0.776
Heptadecenoic	C 17:1	0.44 ^a	0.39 ^b	0.41 ^{ab}	0.010	0.003	0.0001	0.649
Stearic	C 18:0	6.93 ^c	10.23 ^a	8.16 ^b	0.235	0.0001	0.007	0.718
Oleic cis	C 18:1n9c	19.98 ^b	24.57 ^a	24.21 ^a	0.370	0.0001	0.004	0.769
Linoleic trans	C 18:2n6t	0.67 ^c	1.90 ^a	1.22 ^b	0.071	0.0001	0.0001	0.002
Linoleic cis	C 18:2n6c	2.07 ^b	2.66 ^a	2.72 ^a	0.072	0.0001	0.247	0.061
Arachidic	C 20:0	0.05 ^b	0.11 ^a	0.06 ^b	0.008	0.0001	0.019	0.399
Linolenic gamma	C 18:3n6	0.06 ^c	0.13 ^a	0.09 ^b	0.007	0.0001	0.0001	0.966
Eicosenoic	C 20:1n9	0.06 ^a	0.06 ^a	0.04 ^b	0.004	0.0001	0.984	0.0001
Linolenic alfa	C 18:3n3	0.36 ^b	1.50 ^a	0.47 ^b	0.052	0.0001	0.0001	0.001
Conjugated Linoleic–rumenic acid	CLA (c9, t11)	0.36 ^c	0.69 ^b	1.82 ^a	0.081	0.0001	0.070	0.205
Eicosadienoic	C 20:2n6	0.12 ^b	0.14 ^b	0.17 ^a	0.008	0.0001	0.042	0.829
Eicosatrienoic Ω6	C 20:3n6	0.11 ^b	0.11 ^b	0.15 ^a	0.010	0.0001	0.101	0.001
Eicosatrienoic Ω3	C 20:3n3	0.08 ^b	0.10 ^b	0.14 ^a	0.011	0.0001	0.008	0.007
Arachidonic	C 20:4n6	0.15 ^a	0.11 ^b	0.12 ^b	0.005	0.0001	0.010	0.001
Total SFA ¹		70.46 ^a	62.92 ^b	63.31 ^b	0.539	0.0001	0.003	0.534
Total MUFA ²		24.24 ^b	28.47 ^a	28.55 ^a	0.425	0.0001	0.021	0.359
Total PUFA ³		3.74 ^b	7.29 ^a	6.97 ^a	0.208	0.0001	0.002	0.016
n-3 PUFA ⁴		0.41 ^b	1.55 ^a	0.57 ^b	0.053	0.0001	0.0001	0.001
n-6 PUFA ⁵		3.07 ^c	5.02 ^a	4.42 ^b	0.128	0.0001	0.008	0.001
n-6/n-3 PUFA ⁶		7.67 ^a	3.50 ^b	7.32 ^a	0.237	0.0001	0.001	0.015
PUFA/SFA		0.06 ^b	0.12 ^a	0.11 ^a	0.004	0.0001	0.003	0.013
DFA		35.10 ^c	46.12 ^a	43.83 ^b	0.367	0.0001	0.0001	0.363
HSFA		45.68 ^a	36.48 ^b	37.90 ^b	0.535	0.0001	0.0001	0.763
h/H ratio		0.49 ^b	0.76 ^a	0.73 ^a	0.020	0.0001	0.001	0.619
AI		2.94 ^a	1.86 ^b	1.91 ^b	0.062	0.0001	0.002	0.683
TI		1.99 ^a	1.68 ^b	1.60 ^b	0.036	0.0001	0.024	0.081

CON, control diet; LIN, linseed diet; HMP, hempseed diet; SEM, standard error of the mean; ¹ saturated fatty acids, ² monounsaturated fatty acids, ³ polyunsaturated fatty acids, ⁴ omega-3 polyunsaturated fatty acids, ⁵ omega-6 polyunsaturated fatty acids, ⁶ omega 6/omega 3 ratio; DFA = MUFA + PUFA + C 18:0; HSFA = C 12:0 + C 14:0 + C 16:0; h/H ratio = (C 18:1c9 + C 18:2n6 + C 20:4n6 + C 20:5n3 + C 22:5n3)/(C 12:0 + C 14:0 + C 16:0); AI = (C 12:0 + 4 × C 14:0 + C 16:0)/(MUFA + PUFA); TI = (C 14:0 + C 16:0 + C18:0)/(0.5 × MUFA + 0.5 × n6 + 3 × n3 + n3/n6); ^a, ^b, ^c—means in rows marked with different uppercase superscripts significantly differ at $p < 0.001$.

The LIN and HMP diets were associated with a significant decrease ($p < 0.0001$) in the total SFAs (62.92 and 63.31 g/100 g total FAME) vs. the CON diet (70.46 g/100 g total FAME). The butyric (C:4) and caprylic (C:8) acids were slightly increased ($p < 0.05$) for the HMP diet, but the caproic acid (C 6:0) was not influenced by the diet. A decrease was registered for capric acid (C 10:0), an important and specific acid for goat milk, with the

LIN (10.67 g/100 g total FAME) and HMP (11.12 g/100 g total FAME) diets vs. the CON diet (12.09 g/100 g total FAME). The lauric acid (C 12:0), the myristic acid (C 14:0), and the palmitic acid (C 16:0) also registered significant decreases. Only the stearic acid (C 18:0), the most abundant long-chain FA in milk, was increased with both the LIN (10.23 g/100 g total FAME) and HMP (8.16 g/100 g total FAME) diets vs. the CON diet (6.93 g/100 g total FAME).

The LIN and the HMP groups had significantly ($p < 0.001$) reduced proportions of the minor MUFAs like miristoleic (C 14:1) and pentadecenoic (C 15:1); the palmitoleic acid (C 16:1) registered a decrease only with the LIN diet. The total MUFA percentage was increased for both the LIN and HMP groups vs. the CON group. The highest increase in the total MUFA was due to the increase of the *cis*-oleic acid (C 18:1n9c), a major MUFA acid, by values of 24.57 g/100 g total FAME with the LIN diet and 24.21 g/100 g total FAME with the HMP diet vs. 19.98 g/100 g total FAME with the CON diet.

The total PUFA concentration showed a highly significant increase ($p < 0.0001$) for both the LIN and HMP diets compared to the CON diet.

The major n-6 PUFA, *cis*-linoleic acid (C 18:2n6c), similarly increased for both experimental groups (2.66 for the LIN diet and 2.72 for the HMP diet) compared to the CON diet (2.07 g/100 g total FAME). The sum of the n6-PUFA acids with the LIN diet was 5.02 g/100 g total FAME, a higher value than with the HMP diet (4.42 g/100 g total FAME), but both were significantly higher compared to the CON diet (3.07 g/100 g total FAME). The major n-3 acid, the α -linolenic acid ALA (C 18:3n3), was highly significantly ($p < 0.0001$) increased only within the LIN diet (1.50 g/100 g total FAME) compared to the CON diet (0.36 g/100 g total FAME) and HMP diet (0.47 g/100 g total FAME).

The percentage of conjugated linoleic acid (CLA), namely the dominant isomer—rumenic acid, *c9*, *t11*-CLA (or C 18:2n *cis911t*)—registered a significant increase ($p < 0.0001$) with the HMP diet (1.82 g/100 g total FAME) compared to the CON diet (0.36 g/100 g total FAME) and LIN diet (0.69 g/100 g total FAME); also, significant differences were noticed between the LIN and CON diets. This confirms that the presence of dietary oil sources rich in C18:2 and C18:3 acids improves the milk quality.

Health-related indices such as n6/n3, PUFA/SFA, DFA, HSFA, h/H, AI, and TI were also influenced (Table 6). The n6/n3 ratio decreased significantly only for the LIN diet. The DFA index increased significantly for both the LIN (46.12) and HEM (43.83) diets vs. the CON (35.10) diet. Also, the h/H ratio increased for both the LIN and HMP diets, while the HSFA index had important decreases for both the LIN and HMP diets. The linseeds and hempseeds determined the significant decrease in both the AI and TI indices.

3.3. Profile of Fatty Acids in Cheese

The results for cheese (Table 7) were expressed as g FAME/100 g total FAME but also as mg fatty acid/100 g cheese, a parameter which is more relevant to the farmers and consumers.

The linolenic- α acid concentration increased in the LIN diet ($p < 0.0001$) compared to the CON and HMP diets, while for the CLA concentration, the HMP diet registered the highest value compared to the CON and LIN diets.

The FAME profile for the principal classes was relatively similar between cheese and milk. Consequently, the SFA decreased for both the LIN (63.60 g/100 g total FAME) and HMP (62.62 g/100 g total FAME) diets compared to the CON diet (67.80 g/100 g total FAME), the MUFA increased for both groups (27.48, and 28.80 g/100 g total FAME, respectively), and the PUFA increased (7.30, and 7.11 g/100 g total FAME, respectively).

The content of n3-PUFAs compared to the CON diet was highest in the case of the LIN diet, similarly as for milk content. The n6-PUFAs were increased by 1.29 times for the LIN diet and by 1.16 times for the HMP diet. Consequently, the n6/n3 ratio values, similar to those found in milk, exhibited a significant decrease within the LIN diet (3.62) compared with the CON diet (6.88) and the HMP diet (8.00).

Table 7. The FA composition in the goat cheese as g FAME/100 g total FAME (%) and as mg fatty acid/100 g cheese.

Fatty Acids in Cheese		(g FAME/100 g Total FAME)					mg Fatty Acid/100 g Cheese				
		CON	LIN	HMP	SEM	p-Value	CON	LIN	HMP	SEM	p-Value
Butyric	C 4:0	0.04	0.05	0.04	0.007	0.642	3.69	3.60	4.01	0.70	0.900
Caproic	C 6:0	1.36	1.35	1.30	0.074	0.841	120.44	125.27	119.99	9.96	0.912
Caprylic	C 8:0	3.08	3.07	3.05	0.092	0.967	264.65	266.92	260.42	15.0	0.953
Capric	C 10:0	11.50 ^a	10.56 ^b	10.78 ^{ab}	0.238	0.027	1099.81 ^a	963.63 ^b	1076.71 ^{ab}	77.2	0.049
Undecanoic	C 11:0	0.40	0.37	0.36	0.017	0.276	38.68	32.37	33.32	2.21	0.122
Lauric	C 12:0	6.31	5.48	5.32	0.293	0.055	555.08	474.48	518.30	39.2	0.341
Tridecanoic	C 13:0	0.18	0.15	0.15	0.015	0.388	15.31	13.02	13.47	1.20	0.363
Myristic	C 14:0	10.88 ^a	9.63 ^b	9.43 ^b	0.352	0.015	966.24 ^a	852.98 ^b	912.83 ^b	54.3	0.033
Myristoleic	C 14:1	0.64	0.56	0.58	0.037	0.300	61.17	48.73	52.15	3.52	0.061
Pentadecanoic	C 15:0	0.27	0.26	0.25	0.010	0.354	25.43	23.30	22.25	1.31	0.253
Pentadecenoic	C 15:1	0.93	0.93	0.93	0.037	0.998	85.57	80.51	79.38	4.81	0.654
Palmitic	C 16:0	25.41	22.55	23.03	0.825	0.047	2385.47	2010.23	2045.85	140	0.137
Palmitoleic	C 16:1	1.87 ^{ab}	1.80 ^b	2.13 ^a	0.090	0.036	176.90 ^{ab}	165.81 ^b	209.94 ^a	13.7	0.048
Heptadecanoic	C 17:0	0.34	0.33	0.31	0.012	0.272	32.40	30.39	28.80	1.69	0.354
Heptadecenoic	C 17:1	0.38	0.37	0.37	0.017	0.945	34.58 ^a	33.96 ^b	33.35 ^b	1.39	0.008
Stearic	C 18:0	7.97 ^b	9.66 ^a	8.53 ^{ab}	0.432	0.030	696.28 ^b	890.32 ^a	820.62 ^{ab}	53.5	0.046
Oleic cis	C 18:1n9c	21.69 ^b	23.78 ^{ab}	24.74 ^a	0.838	0.046	1939.63 ^b	2240.84 ^a	2232.33 ^a	117	0.050
Linoleic trans	C 18:2n6t	1.03 ^b	1.84 ^a	1.24 ^{ab}	0.199	0.027	82.35 ^b	212.14 ^a	131.10 ^b	16.6	0.0001
Linoleic cis	C 18:2n6c	2.46	2.79	2.85	0.120	0.063	216.26 ^b	257.29 ^a	244.01 ^a	15.2	0.047
Arachidic	C 20:0	0.06 ^b	0.14 ^a	0.07 ^b	0.016	0.001	6.57 ^b	14.91 ^a	7.61 ^b	1.54	0.0001
Linolenic gamma	C 18:3n6	0.12	0.15	0.10	0.020	0.213	11.59	14.05	10.95	1.93	0.514
Eicosenoic	C 20:1n9	0.05	0.04	0.05	0.009	0.815	5.31 ^b	4.03 ^a	3.73 ^a	0.504	0.048
Linolenic alfa	C 18:3n3	0.48 ^b	1.30 ^a	0.47 ^b	0.123	0.0001	43.35 ^b	121.98 ^a	63.04 ^b	11.2	0.0001
Conjugated	CLA (c9, t11)	0.78 ^b	0.74 ^b	1.89 ^a	0.177	0.001	80.50 ^b	68.16 ^b	157.21 ^a	23.7	0.037
Linoleic-rumenic acid											
Eicosadienoic	C 20:2n6	0.13	0.15	0.19	0.019	0.104	13.03 ^b	13.72 ^b	17.04 ^a	1.97	0.032
Eicosatrienoic-n6	C 20:3n6	0.12	0.10	0.13	0.0233	0.536	11.80 ^b	9.12 ^b	13.46 ^a	2.45	0.050
Eicosatrienoic-n3	C 20:3n3	0.10	0.12	0.11	0.025	0.819	8.72 ^b	10.48 ^{ab}	11.15 ^a	2.23	0.047
Arachidonic	C 20:4n6	0.13	0.11	0.13	0.009	0.065	11.82 ^b	9.70 ^a	12.07 ^b	0.943	0.003
Total SFA		67.80 ^a	63.60 ^b	62.62 ^b	1.230	0.087	6210.05 ^a	5701.42 ^b	5864.18 ^b	283	0.016
Total MUFA		25.56 ^b	27.48 ^a	28.80 ^a	0.771	0.021	2303.16 ^b	2573.88 ^a	2610.88 ^a	200	0.008
Total PUFA		5.35 ^b	7.30 ^a	7.11 ^a	0.563	0.024	479.42 ^b	716.64 ^a	660.03 ^a	85.3	0.009
n-3 PUFA		0.58 ^b	1.42 ^a	0.58 ^b	0.158	0.001	52.07 ^b	132.46 ^a	74.19 ^b	12.5	0.0001
n-6 PUFA		3.99 ^b	5.14 ^a	4.64 ^{ab}	0.337	0.039	346.85 ^b	516.02 ^a	428.63 ^b	53.8	0.031
n-6/n-3 PUFA		6.88 ^a	3.62 ^b	8.00 ^a	0.626	0.001	6.66 ^a	3.90 ^b	5.78 ^a	0.572	0.000
PUFA/SFA		0.08 ^b	0.11 ^a	0.11 ^a	0.010	0.022	0.08 ^b	0.13 ^a	0.11 ^a	0.009	0.0001
DFA		38.88	44.44	44.44	1.650	0.031	3478.86	4180.84	4091.53	251	0.005
HSFA		42.60 ^a	37.66 ^b	37.78 ^b	1.390	0.028	3906.79 ^a	3337.69 ^b	3476.98 ^b	216	0.040
h/H ratio		0.59 ^b	0.76 ^a	0.77 ^a	0.046	0.040	0.58 ^b	0.81 ^a	0.75 ^a	0.059	0.009
AI		2.43 ^a	1.91 ^{ab}	1.84 ^b	0.161	0.023	2.45 ^a	1.79 ^b	1.90 ^b	0.188	0.033
TI		2.66 ^a	2.01 ^b	2.21 ^b	0.073	0.024	2.73 ^a	1.93 ^b	2.17 ^b	0.167	0.050

CON, control diet; LIN, linseed diet; HMP, hempseed diet; SEM, standard error of the mean; ¹ saturated fatty acids, ² monounsaturated fatty acids, ³ polyunsaturated fatty acids, ⁴ omega-3 polyunsaturated fatty acids; ⁵ omega-6 polyunsaturated fatty acids, ⁶ omega 6/omega 3 ratio; DFA = MUFA + PUFA + C 18:0; HSFA = C 12:0 + C 14:0 + C 16:0; h/H ratio = (C 18:1c9 + C 18:2n6 + C 20:4n6 + C 20:5n3 + C 22:5n3)/(C 12:0 + C 14:0 + C 16:0); AI = (C 12:0 + 4 × C 14:0 + C 16:0)/(MUFA + PUFA); TI = (C 14:0 + C 16:0 + C 18:0)/(0.5 × MUFA + 0.5 × n6 + 3 × n3 + n3/n6); ^a, ^b—means in rows marked with different uppercase superscripts significantly differ at $p < 0.001$.

The PUFA/SFA ratio was significantly improved in both the LIN and HMP groups (0.11 and 0.11, vs. 0.08 for CON diet, respectively). The content of DFA increased but not significantly in cheese, in a similar way with the milk fat. The HSFA index registered a reduction of almost 12% with the LIN diet and nearly 11% with the HMP diet. The h/H ratio was increased between 1.28–1.30 times for each diet. The value of the AI and TI indices decreased with both LIN and HMP diets.

4. Discussions

Although the energy supply of LIN and HMP diets was higher than the CON diet (due to the replacement of a fat-extracted meal with oilseeds), the milk yield was not statistically different among groups. This might be related to the high proportion of dietary fat in LIN and HMP groups. This lack of effect is in line with the results of two previous studies [9,15], where the inclusion of 9.3% DM of linseeds or hempseeds in the diet for Alpine lactating goats also resulted in similar milk yields among groups.

The increased milk fat content is consistent with the fact that replacement of sunflower meal with linseeds and hempseeds represents a supplementation of total dietary lipids,

equivalent with 4.6% of DM intake. The effect of increasing milk fat after adding linseed to the diet was also reported in ewes by [7], in Cilentana grazing goats by [26], and by [9] in Alpine goats for both oilseeds. A similar effect of the hempseeds on the milk fat content was observed by [16], also on ewes, along with an increase of the milk yield. Even in iso-energetic diets, Sampelayo et al. [27] found that the replacement of starch with fat, as energy sources, has increased milk fat content and stimulated the presence of oleic, vaccenic, and rumenic acids, as well as linolenic acid and other trans fatty acids in goat milk.

Whereas the protein and casein contents were not modified by the LIN and HMP diets, the lactose content was higher in these diets. This increase was associated with the higher energy levels in these diets (0.865 UFL and 0.863 UFL compared to 0.787 UFL). The increased lactose levels could also be attributed to a higher glucose availability for lactose synthesis in the mammary gland as a result of feeding with these lipid-enriched diets. This effect was presented by other researchers [24] in goats fed starch-enriched or lipid-supplemented diets. Tudisco [8] also reported an increase in lactose content for goat fed linseed (4.61%) compared with pasture-fed goat (4.57%).

The milk urea-N registered a decrease for both LIN and HMP diets; this decrease is also in line with other authors [28] who showed that milk urea nitrogen is also negatively correlated with the increasing energy content of the diet, like in our study for LIN and HMP groups.

The levels of short SFAs in milk observed in our study were slightly decreasing. They enhance the milk's taste and flavor and, according to Chilliard [4], the SFAs serve as an energy source for the proper functioning of internal organs, the nervous system, and muscles in the human body.

The decrease of the levels of long SFAs was a natural consequence of dietary long-chain FA, as presented by [4].

Other researchers [13] reported similar results, such as a decrease in SFAs *de novo* synthesized (C 10:0-C 16:0) and an increase in C 4:0 and C 18:0 acids, as well as in PUFA amounts in goat milk, after the dietary inclusion of hempseed oil. Similar results were obtained by [10] concerning elevated stearic acid (C 18:0) levels in Alpine lactating goats fed with a diet comprising 9.4% hempseed in DM, wherein they also noted a reduction in the C 8:0-C 16:0 acids.

The magnitude of the increase of the concentration of total milk MUFA percentage was comparable in both the LIN and HMP groups. This increase also contributed to the significant increase in the DFA index (46.12 for the LIN diet and 43.83 for the HMP diet vs. 35.10 for the CON diet).

The increase in *cis*-linoleic acid in milk was due to its prevalence in hempseeds (53.41%) and its significant presence in linseeds (16.42%). This effect was presented by other authors [29] who observed that the sum of oleic, linoleic, and α -linolenic acids in the milk fat exhibited a linear increase corresponding to the dietary daily intake of unsaturated lipids. The increased presence of α -linolenic acid exclusively in the milk from the LIN diet emphasizes the effect of the linseeds on the milk composition by the presence of this major FA in the linseeds in a very high amount, 51.33%. A similar effect was presented by [9]. The difference in the major PUFA composition in these two oil seeds is summarized in the n6/n3 ratio, for milk which significantly decreased only with the LIN diet but remained unchanged within the HMP diet due to the absence of an increase in milk n3-PUFAs.

Our obtained values for the LIN and HMP diet on CLA isomers are in line with other studies on various diet types and oilseeds. Correddu et al. [30] found increased levels of vaccenic acid in milk of dairy sheep fed linseed. High levels of 0.7% for both CLA isomers (rumenic acid and *t*10 *c*12 CLA) and for oleic acid were reported by [7] for ewes fed extruded linseeds. Increased levels of CLA isomers were also found for diets with hempseed as presented by other authors [14] on Carpathian goats and by [10] on Alpine lactating goats supplemented with 9.4% hempseed in different types of diets (based on pasture hay/shrubs–grass rangeland/less fermentable or less degradable ingredients). According to Cozma et al. [13], a very high increase of CLA-rumenic acid concentration

in milk fat was observed when goats' diets were supplemented with hempseed oil. Our CLA values for HMP diet were similar to those from an extensive, grazing-based, milk production system presented by the author Slots [31], who found that cows fed natural pasture yielded a CLA content of 1.75%.

Both vaccenic and rumenic acids are known for anti-atherosclerotic effects by lowering blood levels of triglycerides and LDL cholesterol fractions, anti-cancer effects, and beneficial effects on the immune system [32,33].

The main fatty acid classes showed comparable values in both cheese and milk; however, only the Cis-linoleic acid registered a high increase in milk, whereas in cheese, the increase was not statistically significant. Also, the CLA-rumenic acid corresponding to the LIN diet was not increased in cheese compared to the CON diet (although significantly different values were observed for milk). However, the direction of changes between LIN or HMP comparing to the CON diet were similar to those registered in milk; therefore, the minor inconsistencies between milk and cheese were caused by the cumulated individual errors (sampling, analyses, etc.).

The health indices were similar for cheese and milk (as FAME), and after expressing FA in an absolute amount (mg FA/100 g cheese), the modifications of the indices were even better highlighted. These indices are consistent with the findings of [30], who recorded decreased values of AI and TI and an increased h/H ratio when dairy ewes were fed linseed. Similar improved health-promoting indices were registered by other authors [34] for semi-hard cheese made from milk of French Alpine goats fed a basal diet with 90 g/kg DM extruded linseed. The results for the n6/n3 ratio, for milk and for cheese, were around 5.0, which is recommended to be below 5.0 in the human diet in order to mitigate the risk of developing cardiovascular diseases and cancer [35].

5. Conclusions

The replacement of sunflower meal with equal quantities of linseeds or hempseeds in the diets of Murciano-Granadina goats, at a level of 11.5% proportion of the total dry matter intake, had no influence on raw milk yield or milk protein content but led to a significant increase in the milk fat content.

In the case of raw milk composition, the inclusion of either LIN or HMP led to the decreases in the SFA proportion and the increases in the MUFA proportion (mainly the oleic acid). Inclusion of LIN induced a significant decrease in the n3:n6-PUFA ratio.

Also, in the LIN diet milk, the ALA (C 18:3n-3) increased by 4.1 times, while the LA (C 18:2n6c) also increased by 1.3 times. In the HMP diet milk, there was no increase in ALA, and the increase of LA was similar with the LIN diet.

The increased contents of healthy FA, such as n3-PUFA, and CLA-rumenic led to higher health indices of the milk corresponding to the goats that were fed a linseed or hempseed diet.

The improved FA profile of the milk collected from the goats fed either LIN or HMP diets was found also in the cheese manufactured from this milk, which is a good base for obtaining premium-labelled dairy products through application of feeding strategies that covers also the non-grazing periods or production systems.

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