INCORPORATING ESSENTIAL FATTY ACIDS IN SANA WITH FISH OIL ADDED

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Abstract

In this study we have determined the optimal concentrations of fish oil added to raw milk on which the optimal embedding fish oil globule of fat in the product. to this end was made from sheep's milk homogenization temperature of 70 ° c and pressure of 200 bar. The fat globule membrane scission occurs when fat molecule occurs accession fat fish oil to milk fat globules and restoration, which is the number greater use of milk protein composition. we used sheep milk and were analyzed three most important essential fatty acids: linoleic acid, linolenic acid and γ-linolenic acid. Fatty acid analysis was performed by gas chromatography and statistical analysis method of embedding threshold comparison method roc curve (receiver operator characteristic = operating characteristic) and have obtained the following theoretical threshold values potting: linoleic acid, not potting reaches concentrations up to 0.15% fish oil added to milk; 0.650% linolenic acid; 1.319% γ-linolenic acid.

Key words: fish oil essential fatty acids

INTRODUCTION

Consumer interest for products in dairy goats and sheep, especially traditional ones is increasing (Park at Haenlein, 2006). Sheep milk is used in the production has biologically important qualities for man with role in lowering body fat (Versiani and others, 2008). It is known that sheep milk is important not only for liver metabolism but also to cover the manganese is used as food for recovery after illness. It is known that people who practice grazing in several regions of the world have a higher life expectancy (Hanreich et al, 2008).

Because fermentation occurring were identified in products are made up of lactic acid bacteria Enterococcus faecalis (Muguerza, B, 2006), Lactococcus lactis subspecies cremoris (Gobbetti, M, 2000), bioactive peptides acting as enzyme inhibitors (ACE) that influence cardiovascular protection body blood pressure reduction, etc.

High biological value products from sheep's milk is given and fat composition. For this purpose we studied sheep milk in terms of composition of essential fatty acids. Approximate composition of sheep milk fatty acid is the saturated FA (SFA) in milk fat level is quite high (More Than 60%) while monounsaturated FA (MUFA) and polyunsaturated FA (PUFA) has approximately 28% and 6%, respectively (Cabiddu et al.,

2003, 2005). Concentration in sheep milk fatty acids is influenced by many factors: genetics, diet, age, number of lactations, lactation period (A. Cartaica, 2008). Depending on the stage of lactation (Daniel blackbird, 2011), the number of lactations (Mierliță D, 2011) essential fatty acids concentration increases with advancing during these periods. Also this component is used to increase the protein and energy rich food (Mierliță D., 2009). Essential fatty acids ω-3 and ω-6 is very important for the human body: the inflammatory and cardiovascular diseases (ShuWanga, 2009), hepatoprotective (Mohamed Makni, 2010), nerone diseases as: attention disorders in children (Nathan Gadoth, 2008), Parkinson's disease (M. Bousquet, 2009), depression (Sabine Riemer, 2010).

MATERIAL AND METHOD

Sana was from sheep's milk collected during the first period of lactation. It was used from 3000 sheep milk mixture. Lactic culture was used to manufacture Ch Hansen, type CHN 11. In raw milk has been introduced fish oil (purchased from the company "Hofigal") and the mixture was homogenized at 70 ° C and a pressure of 200 bar. in three steps. Next to the classic dupe manufactured sana (Costin GM, 2005, 2007). Depending on the concentration of fish oil in raw milk samples were coded as follows: blank without fish oil: S_0; with the addition of 0.05%: $S_{0,\ 05}$; with the addition of 0.10%: $S_{0,\ 10}$; and with the addition of 0.015%: $S_{0,\ 15}$.

Sensory analyzes were performed by five unauthorized persons.

Physical and chemical analyzes were performed: determination of acidity (according to ISO 6091/2008), the percentage of fat (according S.T.A.S. 6352/2-87).

For fatty acid analysis were performed gas-chromatographic determinations and were detected 19 fatty acids. Working protocol for gas-chromatographic analysis was: milk fat was extracted by using the following protocol: about 1ml of milk samples were mixed with 0,6 ml ammonia 25%, 2ml EtOH, 4ml Ethyl ether and 4 ml hexane and then agitated for 2-3min. After this process the lower layer (the ammonia layer) was discarded. Following this step the mixture was passed through a cellulose filter with Na_2SO_4 and then brought to dryness; transesterification: fatty acids were converted to methyl esters by reaction with boron trifluoride/methanol at 80ºC for two hours in a closed Pyrex glass tube. The content was transfered into a separatory funell; the methyl ester extraction: the extraction was made using 10 ml hexane, the hexanic fractions collected were dried using anhydrous sodium sulfate, filtered, concentrated under a nitrogen stream and finally re-eluted in 1 mL hexane. Fatty acids were analyzed by gas chromatography (GC) with flame ionization detection (FID). A 1µL sample was injected into the Shimadzu GC-17A series gas-chromatograph, equipped with a 30m polyethylene glycol coated column (Alltech AT-WAX, 0.25mm I.D.,

0.25µm film thickness). Helium was used as the carrier gas at a pressure of 147 kPa. The injector and detector temperatures were set at 260ºC. For the oven temperature the following program was used: 70ºC for 2 min. then raised to 150ºC at 10ºC/ min. rate and held at 150ºC for 3min., then further raised up to 235ºC at a 4ºC/min; fatty acids were analyzed by gas chromatography (GC) with flame ionization detection (FID). A 1µL sample was injected into the Shimadzu GC-17A series gas-chromatograph, equipped with a 30m polyethylene glycol coated column (Alltech AT-WAX, 0.25mm I.D., 0.25µm film thickness). Helium was used as the carrier gas at a pressure of 147 kPa. The injector and detector temperatures were set at 260ºC. For the oven temperature the following programe was used: 70ºC for 2 min. then raised to 150ºC at 10ºC/ min. rate and held at 150ºC for 3min., then further raised up to 235ºC at a 4ºC/min.

Statistical analysis of data for comparison of samples has been used ANOVA, Tukey and Duncan tests and to compare with blank Dunnet test. To determine the optimal amount of fish oil was used comparing ROC curves (Receiver Operator Characteristic = Operating Characteristic) and Fourier analysis of the correlation type (Sipos, C. et al 2004; Teuşdea A, et al, 2008; Teuşdea A., et al, 2008; Teusdea A, 2009).

RESULTS AND DISSCUSIONS

Sensorial in sana samples without and with fish oil did not taste and smell of fish observed after 7 days at refrigerator temperature maintenance or other foreign taste and odor. In terms of physical and chemical analyzes are not significant differences between samples.

Table 1.

Multiple comparisons for samples S_0; $S_{0.05}$; $S_{0.10}$; $S_{0.15}$ sana by Tukey algorithm (HSD) / 95.00% for fatty acids detected chromatographic

Fatty acids	$S_{0.05}$~S_0	$S_{0.10}$~ S_0	$S_{0.15}$ ~S_0	$S_{0.05}$ ~$S_{0.10}$	$S_{0.05}$ ~$S_{0.15}$	$S_{0.10}$ ~$S_{0.15}$
Caproic	0.00223	0.00223	0.00223	1.00000	1.00000	1.00000
Caprilic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Capric	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Lauric	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Miristic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Miristoleic	1.00000	0.22207	0.00223	0.22207	0.00223	0.00223
Pentadecanoic	0.00223	1.00000	0.00223	0.00223	0.21490	0.00223
Cis-10 pentadecanoic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Palmitic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Palmitoleic	0.00223	0.00223	0.00223	0.00223	0.00223	0.01436
Heptadecanoic	1.00000	0.52570	1.00000	0.53484	1.00000	0.52570
Cis-10 heptadecanoic	0.00223	0.19072	0.19072	0.00434	0.00434	1.00000
Stearic	0.00223	0.14908	0.00223	0.00223	0.00223	0.00223
Oleic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Elaidic	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
Linoleic ω6	0.00485	0.00485	0.00485	0.00485	0.00485	0.00485
Linolenic ω3	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223
γLinolenic ω6	0.00223	0.00223	0.00223	0.00223	0.00223	0.00223

To validate the statistical variations in samples sana fatty acids as conclusion of all fatty acids detected in the product chromatographic analysis were performed by ANOVA test for multiple comparisons. Table 1 shows these results for centralization and Table 2 Tukey algorithm for iterative algorithm Dunnet.

Table 2.

Multiple comparisons for samples S_0; $S_{0.05}$; $S_{0.10}$; $S_{0.15}$ by sana
by Dunnet algorithm / 95.00% for the chromatographic fatty acids detected.
Critical range test Dunnet: [-2.89061, +2.89061]

Fatty acids	$S_{0.05} \sim S_0$	$S_{0.10} \sim S_0$	$S_{0.15} \sim S_0$
Caproic	-171.04719	-171.04719	-171.04719
Caprilic	-160.08331	-80.04166	-253.03491
Capric	-252.48038	-76.50921	-172.14571
Lauric	-152.02796	-38.89087	-49.49747
Miristic	28.18855	-20.13468	10.06734
Miristoleic	0.00000	2.13201	-12.79204
Pentadecanoic	10.78328	0.00000	12.93993
Palmitic	237.76806	18.54928	97.80530
Palmitoleic	43.16689	8.22226	12.33340
Heptadecanoic	0.01696	1.41335	0.00000
Cis-10-heptadecanoic	8.98177	2.24544	2.24544
Stearic	-12.12678	2.42536	104.29032
Oleic	249.20798	49.41193	81.63710
Elaidic	23.23790	15.49193	-25.81989
Linoleic (ω6)	24.82172	56.27038	143.23368
Linolenic (ω3)	37.94733	79.05694	94.86833
γ-Linolenic (ω6)	29.39874	53.45225	98.88666

The analysis of surface samples sana chromatograms and fish oil resulting increase their values with increasing concentration of fish oil samples, which shows that the percentage of fatty acids to total fatty acids in the samples increases on addition of fish oil, mainly (Table 3).

Table 3.

Comparison of surface chromatography (ua)
of essential fatty acids in sana

$S_{0,05}$	S(u.a.)	$S_{0,10}$	S(u.a.)	$S_{0,15}$	S(u.a.)	Ulei_peste	S(u.a.)
Linoleic	41.9709	**Linoleic**	43.8620	**Linoleic**	43.6881	**Linoleic**	64.2705
Linolenic	12.9837	**Linolenic**	16.1073	**Linolenic**	15.0795	**Linolenic**	178.0618
γ-Linolenic	15.2309	**γ-Linolenic**	13.5434	**γ-Linolenic**	12.8500	**γ-Linolenic**	40.2639

ROC curves use genuine comparison method, we determined the optimal threshold potting essential fatty acids in sana: Figure 1, linoleic acid; Figure 2, linolenic acid; Figure 3, γ-linolenic acid.

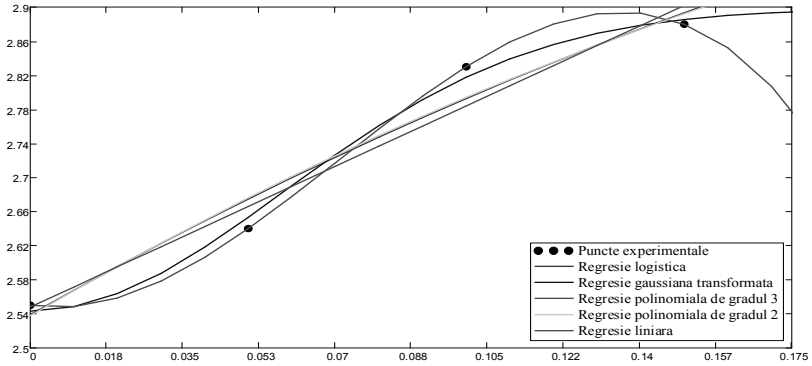

Figure 1. Regression functions studied in the case of linoleic acid of sana

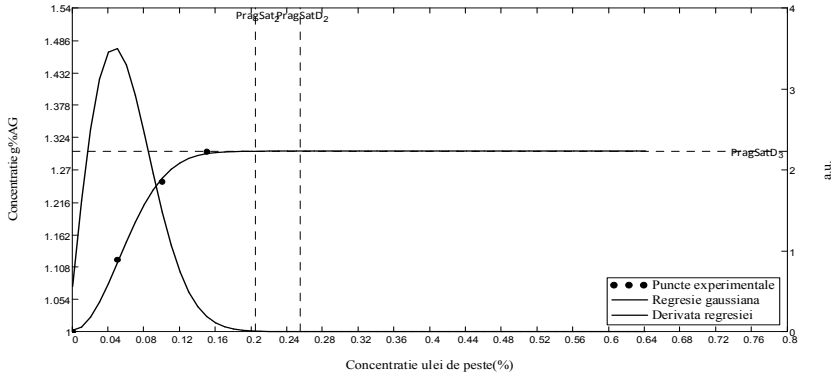

Figure 2. Asymptotic threshold determination linolenic acid potting in sana, with transformed gaussian function

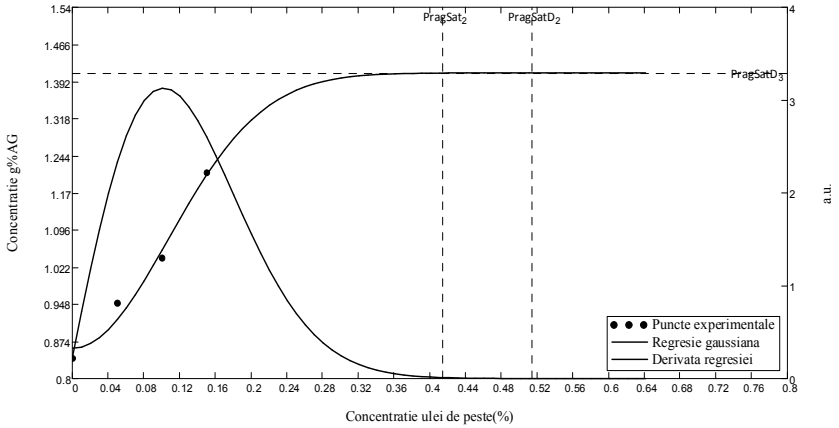

Figure 3. Asymptotic threshold determination potting γ-linolenic acid in sana, with transformed gaussian function

Figure 1 shows that the addition of up to 0.15% fish oil in the milk of manufacture sana potting limit is not reached linoleic acid in fat globule of product. Embed range linolenic acid in fat globule sana is between 0.2 ÷

0.25% (Figure 2), fish oil added to milk and γ-linolenic acid is between 0.4 ÷ 0.5% . That if linoleic acid is not reached maximum potting may be due to the fact that homogenization was performed first milk and fish oil, followed by pasteurization it is possible that high-fat globule membraba to heat and thus split pass some of the fat globule in plasma.

After centralizing embedding threshold values of fatty acids in fat globule (Table 4), we see that they differ not significantly depending on the function used.

Table 4.

Asymptotic threshold values potting essential fatty acids from sana

Fatty acids	Embedding threshold (from regression values)	Embedding threshold (the derivative regression values)	Embedding threshold (theoretical)
linoleic	threshold is not reached maximum embedding up to 0.15%		
linolenic	1.301	1.301	0.650
γ-linolenic	1.409	1.409	1.319

By way ANOVA, Tukey test to determine the optimal concentration of fish oil that includes in sana fat globule, according to data.

Table 5.

Fatty acid concentrations to a maximum of embedding the samples sana

Fatty acids	Conc. of fish oil(%)	Type asymptotic threshold
Linolenic	1.3009	din valorile regresiei
Linolenic	1.3011	din valorile derivatei regresiei
Linolenic	0.6505	teoretic
γ -Linolenic	1.4090	din valorile regresiei
γ -Linolenic	1.4093	din valorile derivatei regresiei
γ -Linolenic	1.3200	teoretic

Table 6.

Number values, sample averages, standard deviation

Variable	No. value	No. valid values	No. invalid value	sum of weights	MD	SD
Conc. Fish oil (%)	6	6	0	6	1.232	0.289

Table 7.

Multiple comparisons of levels of essential fatty acids sana to embed the maximum produced by Tukey algorithm (HSD) / 95.00%.
Tukey critical Vloarea statistics: 3927.

Categories	Differences	Differences standardized	Critical value	Pr. > Dif.	Significantly

γ-Linolenic ~ Linolenic		0.295	1.349	2.776	0.249	Nu

Categories	MD	Groups
γ-Linolenic	1.379	A
Linolenic	1.084	A

It is noted that there are significant differences between potting optimal thresholds, so they can consider the average concentration, 1.232% fish oil added in milk, to the theoretical optimal limit potting fish oil in sana.

CONCLUSIONS

The research conducted has shown that sana may be made from sheep's milk with added fish oil. The recommended concentration in terms of technology, is 0.15% lower than that resulting from theoretical view points. Further studies are needed to remove the risk of passing the fat globule fatty acids in plasma by high temperature pasteurization of milk, followed by maintaining the freezing temperatures in order to rehydrate protein and fat cell membranes recovery, possibly split heat, followed by mixing milk with added fish oil. Is necessary and deodorizing mixture of milk with fish oil.

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