Progress in Nutrition 2022; Vol. 25, N.2: e2023021 DOI: 10.23751/pn.v25i2.13608

Original article

Fatty acid composition, lipid quality indices, and textural profile of traditional Kargı Tulum PDO cheese

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Abstract. The purpose of the study is to examine the effects of fatty acid profile, texture profile, and important lipid nutritional health indices which are atherogenic index(AI), health promoting index(HPI), thrombogenicity index(TI) and Hypocholesterolemic /Hypercholesterolemic ratio (H/H) on geographically indicated Kargı Tulum cheese produced from cow, sheep, and goat milk. According to our results obtained in the study, polyunsaturated fatty acids (PUFA) ratios of goat cheeses (2.386%) were found to be significantly higher ($p< 0.05$) than cow and ewe cheeses (0.539% and 1.001%, respectively). In the study, the AI value of cow cheese was found to be 3.65, significantly higher ($p<0.05$) than that of sheep and goat cheeses. TI value was found to be the highest in cow cheese i.e.,4.1405.

Key words: Lipid quality indices; fatty acid profile; Kargı Tulum cheese

Introduction

Cheese is very important in terms of human diet due to the variety of fatty acids it contains in the structure of milk fat. From a nutritional point of view, the digestibility of fat in cheeses is quite high (1). In addition, cheese is considered a part of a healthy diet because it is an important source of short chain fatty acids (2). The milk fat in the cheese contains saturated fatty acids (SFA) such as myristic and palmitic acids, which increase the blood plasma cholesterol level and increase the incidence of coronary heart diseases. However, cheese contains monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA), which have a positive effect on health, in its structure in varying proportions according to cheese types and the processes applied to milk, and it also supports the effect of reducing the risk of cardiovascular diseases (3,4-6). In addition, consuming dairy products with a low health lipid indices has a decreasing effect on total cholesterol and the LDL-cholesterol in human blood plasma (7,8). Regular consumption of cold-water fish and therefore long-chain omega 3 fatty acids (C18:3 ; ALA) reduces the incidence of death such as sudden death, myocardial infarction, and stroke.

Fatty acids in our diet are generally thought to have positive or negative preventive and therapeutic effects against diseases. The structure of fatty acids consists of a mixture of saturated fatty (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA), so it is necessary to determine their nutritional and therapeutic values (8). It is important that the fatty acids contained in the milk fat structure turn into flavour compounds during the ripening of cheese and are the precursors of many volatile flavour components. In addition, proteolysis is an important biochemical pathway in which the proteins at the ripening stage of cheese are broken down into amino acids and short peptides, and it is a process that directly affects the flavour development of cheese (9). At this stage, the enzymatic hydrolysis of lipids is carried out by lipolytic enzymes, especially

the lipase enzyme. Lipases occur naturally in milk or can be produced by microorganisms. In the formation of flavour in cheese, fatty acids formed because of the hydrolysis of milk fat and secondary products resulting from the breakdown of fatty acids significantly affect the aroma of the cheese. An important factor in the characteristic aroma of Kargı Tulum, which is a type of cheese produced without fat, is that it is a type of cheese with a high milk fat ratio. Kargı Tulum cheese is a type of cheese with high acid character, milk fat content, produced by using low amount of rennet in traditional production method and using low temperature long-term incubation (15-18 ºC 24 hours) used for coagulation. In addition, Pike Coverall, which has a yellowish colored appearance due to its high oil content, has a semi-hard textural structure. Due to these qualities, it has an aroma that makes you feel the difference from other Tulum cheeses by leaving an oily and sharp taste-odor in the mouth. Considering the above-mentioned criteria, it is seen that Kargı Tulum cheese has qualities originating from the plants in the geography in the feeding of the animals from which the milk used in its production is obtained, in terms of taste and smell.

Tulum cheese is the most famous and widely consumed cheeses among the matured cheeses in Turkey. The name Tulum comes from the animal skin bag in which the cheese is placed for the ripening period. Milk fat is one of the main components in the quality of Tulum cheese. It is well established that milk fat is essential for development of correct flavour in cheese during ripening (10).

Traditional products have specific characteristics that differentiate from other similar products. Because of non-use of any additives in their production process and their beneficial effects on human health (11). Kargı Tulum Cheese is the preferred traditional Turkish cheese with its unique characteristic taste and aroma, produced from cow, sheep, and goat milk and matured in bags overalls by the traditional cheese masters of Kargı in the northwest Black Sea region, which has a Protected Designation of Origin (PDO) status. It is seen that Kargı Tulum cheese, which has a high ALA level, is important in terms of nutrition and heart protective effect. The aim of the study is to reveal the fatty acid profile, texture profile and nutritional and health lipid indices that characterize Kargı Tulum cheese. Due to the scarcity of studies in this area, there is not enough information about the structure of the Kargı Tulum overall.

Materials and methods

Kargı tulum cheese is a type of tulum cheese that starts at the end of spring, coinciding with the breeding period of sheep from raw milk using traditional methods of sheep, goat, and cow milk, produced in the plateaus of the region at an altitude of 1500-1850 altitude during the summer months and consumed after maturing in the plateau. Kargı Tulum cheese is produced in small home-type enterprises using traditional methods. Fresh cheeses produced daily are firstly packed in 15-20 kg cloth bags and stored in a room at 15-18 C. After the cloth bags are filled, the cheeses are pressed into larger bags with a capacity of 50 kg in the same way as not to take air as much as possible. These bags, on the other hand, are matured for a period of 4-5 months while the bags are renewed every 15-20 days during the summer months. After the traditional production process, cheeses are put on the market after being filled with a packaging technique specific to the region in 500 grams, 1 and 1.5 kilograms produced by processing sheepskin. In this research Kargı Tulum cheeses which is produced raw sheep, goat and cow milk ripened for four months in wells located in Kargı were taken from its traditional producers by Kargı municipality and brought to our laboratory in their own packages. Samples were kept under refrigerator conditions at +4 °C, until being analysed.

Chemical analysis

Total solids (TS) content of the cheese samples was measured by gravimetric method, fat was determined by the Gerber method, salt was measured by Mohr titration, acidity was determined and ash content was determined using the method of Kurt et al. (12), pH was measured using a pH meter (model Adwa AD1000) with combined glass electrode.

Fatty acid profile

The lipids were dissolved in 5 mL of hexane for conversion to fatty acid methyl esters (FAME's). The fats, the FAME composition analyses were carried out using a Thermo Scientific brand gas chromatography device with a flame ionization detector and a Restek RTX-2330 capillary column (30 m, 0.25 mm id, 0.2 μm film thickness). The sample injection volume was 1 μl. The column furnace temperature was set at 250 °C to increase at 4 °C/min and remain constant at 100 °C for 3 minutes at and the time to reach this temperature was programmed to be 17 minutes. The injection temperature was set to 250 °C, the detector temperature was set to 260°C, and Helium was used as the carrier gas.

Health lipid indices

Short chain saturated fatty acid (SCFA), and medium chain saturated fatty acids (MCFA) were calculated according to following equations:

$$SCFA = C_{4:0} + C_{6:0}$$

$$MCFA = C_{8:0} + C_{10:0} + C_{11:0} + C_{12:0} + C_{13:0} + C_{14:0} + C_{15:0}$$

The atherogenicity index (AI) and thrombogenicity (TI) indices were calculated, as proposed by Ulbricht and Southgate (1991) through the equations (3, 8):

$$AI = (C_{12:0} + (4 \times C_{14:0}) + C_{16:0}) / \Sigma UFA$$

$$TI = (C_{14:0} + C_{16:0} + C_{18:0}) / ((0.5 \times \Sigma MUFA) + (0.5 \times \Sigma n\text{-}6\ PUFA) + (3 \times \Sigma n\text{-}3\ PUFA) + (n\text{-}3/n\text{-}6))$$

Hypocholesterolemic Fatty Acids (DFA)

$$DFA = \Sigma MUFA + \Sigma PUFA + C_{18:0}$$

Hypercholesterolemic Fatty Acids (OFA)

$$OFA = C_{12:0} + C_{14:0} + C_{16:0}$$

Hypocholesterolemic/Hypercholesterolemic ratio (H/H) was calculated according to the fatty acids composition using the following formula (13).

$$HH = (DFA) / (OFA)$$

The texture profile analysis

The texture profile analysis (TPA) was performed at room temperature using a texture analyser (TA-XT Plus) with a 5 kg load cell. The Tulum cheeses were cut into 25 mm cubes. The TPA conditions were under stainless compression probe (5 cm diameter) as follows: pre-test speed 1.0 mm/s, test speed of 5.0 mm/s. Texture profile analysis (TPA) was performed by compressing the sample to 40% of its original height, and 5 s between two compressions. The values for hardness(N), adhesiveness (g.s), cohesiveness, springiness, chewiness(N) and gumminess(N) were determined according to texture analyser manual.

Statistical analysis

One way analysis of variance (ANOVA) was done to establish statistical differences between the chemical, microbiological, and textural properties of the samples. Tukey multiple range test were applied to determine differences among different groups using SPSS for Windows (version 25).

Results and discussions

The results of the chemical analyses applied to the cheese samples (titratable acidty, total solids, fat, salt, protein, ash) are shown in Table 1. The results of the chemical analysis measurements in the samples are in agreement with other researchers (12-15). There were no significant differences between the cheese samples (cow, sheep and goat) in the case of compositions.

According to the literature data, the fatty acid composition in milk fat depends on many different factors such as season, breed, lactation stage, age and geographical location (13). Fatty acids prevent atherosclerosis and coronary thrombosis because of

Table 1. Some chemical composition of Kargı Tulum cheese(%)*

Samples	Dry matter ± SD	Fat ± SD	Salt ± SD	Ash ± SD	Lactic acid ±SD	Protein ± SD
Cow cheese	69,54 ± 4,88^{a}	31,5 ± 2.29^{a}	3,03 ± 1.17^{b}	3,15 ± 2,11^{a}	0,89 ± 0.85^{a}	24,01 ± 2.25^{b}
Sheep cheese	67,16 ± 7,35^{a}	36,5 ± 3.74^{b}	3,87 ± 1.25^{c}	4,52 ± 1,45^{b}	0,82 ± 1.12^{a}	19,08 ± 3.07^{a}
Goat cheese	70,64 ± 5,23^{a}	33,0 ± 6.41^{ab}	2,56 ± 0.97^{a}	2,70 ± 2.05^{a}	0,88 ± 0.80^{a}	22,04 ± 3.67^{b}

*Means in the same column followed by different letters represent significant differences($p<0.05$). Data are expressed as mean ± standard deviation.

their effects on serum cholesterol and low-density lipoprotein-cholesterol concentration. So, fats not only provide energy in the diet, but also have an important role in promoting good health (17,18) In our study, twenty-nine fatty acids were determined under experimental conditions in cheese samples seventeen of which were saturated fatty acids and twelve were unsaturated fatty acids and the results are given in Table 2. Due to the limited information of fatty acid profile of Kargı Tulum cheese, the results obtained in this research were discussed with different lipid quality indices. The fatty acid compositions in the cheese samples were qualitatively similar, especially in terms of saturated fatty acids. In all the analysed cheese samples, saturated fatty acids (SFA) were dominant. Ewe cheese had significantly lower ($p< 0.05$) contents of these acids than others. The most abundant saturated fatty acids in all samples were palmitic acid, stearic acid, and myristic acid with the levels of 27.39-39.82, 8.21-15.88, and 9.27-13.13 % respectively, but capric acid was found to be considerably higher only in goat milk cheese compared to ewe and cow milk.

Milk fat has high concentrations of short chain fatty acids compared with other foods, so it is the main source of SCFA in the diet. SCFA's are important from a nutritional point of view. These fatty acids have the characteristic of being volatile, which will give much of aroma and flavour of cheese (19,20) Short chain fatty acids (SCFA) (C_4 to C_{10}), were found to be slightly higher in goat cheese sample than in cow and ewe cheeses, indicating that lipolysis is higher in goat cheese. In the same table the short chain fatty acids appear in appreciable concentrations and present significant differences ($p<0.05$) between the cheese samples. Considering this group of fatty acids the highest contents of butyric acid ($C_{4.0}$) were found in the samples from cow cheeses. It has been determined that the results obtained are in accordance with the results of other studies on this subject (7,11,22). Short chain fatty acids are important in terms of their positive effects on human health, such as the anti-inflammatory activity of butyric acid and its protective effects from colorectal and breast cancers (13). Capric acid ($C_{10.0}$), another important short chain fatty acid, is the highest in goat cheese sample and its ratio in total fatty acids was determined as 8.7%.

The health lipid indices of cheese samples are presented in Table 3. Lipid health indices allow us to have more information about the nutritional and health effects of consumption of milk and dairy products. In this context, some researchers focus on the determination of lipid health indices such as DFA, AI, TI, SCFA and H/H as well as fatty acid composition in cheeses. In this respect, low AI, TI and H/H values as well as high DFA are desired values (5). The difference in the amount of monounsaturated fatty acids (MUFA) in the cheese samples was found to be statistically significant ($p< 0.05$), and it was determined to be significantly higher in sheep cheese than cow and goat cheeses. According to our results obtained in the study, polyunsaturated fatty acids (PUFA) ratios of goat cheeses (2.386%) were found to be significantly higher ($p< 0.05$) than cow and ewe cheeses (0.539% and 1.001%, respectively). In a study conducted by Prandini et al.(23), the amount of SFA in cow cheese was found to be between 65.23% and 68.52%, while the amount of MUFA was between 27.90% and 31.19% and the amount of PUFA was between 3.48%

Table 2. Fatty acid content of Kargı Tulum cheese (%)*

Fatty Acid	Cow cheese	Sheep cheese	Goat cheese
C4:0	1,160 ± 0.08[b]	1,014 ± 0.07[a]	0,892 ± 0.07[a]
C6:0	1,240± 0.12[b]	0,838 ± 0.05[a]	1,549 ± 0.02[c]
C8:0	0,937 ± 0.07[a]	0,806 ± 0.02[a]	2,228 ± 0.03[b]
Cl0:0	2,587 ± 0.11[a]	2,675 ± 0.09[a]	8,726 ± 0.11[b]
C11:0	0,043 ± 0.09[b]	0,000 ± 0.00[a]	0,049 ± 0.05[c]
C12:0	3,523 ± 0.05[b]	2,205 ± 0.07[a]	3,560 ± 0.01[b]
C13:0	0,097 ± 0.02[b]	0,064 ± 0.01[a]	0,065 ± 0.07[a]
C14:0	13,139 ± 0.07[b]	9,701 ± 0.11[a]	9,272 ± 0.09[a]
C14:1	0,681 ± 0.04[b]	0,667 ± 0.08[b]	0,388 ± 0.03[a]
C15:0	1,591 ± 0.03[b]	1,426 ± 0.03[b]	1,074 ± 0.02[a]
C15:1	0,417 ± 0.02[c]	0,323 ± 0.01[b]	0,227 ± 0.07[a]
C16:0	39,822 ± 0.17[b]	27,395 ± 0.09[a]	29,995 ± 0.14[a]
C16:1	2,120 ± 0.12[c]	1,636 ± 0.05[b]	0,893 ± 0.07[a]
C17:0	0,898 ± 0.07[a]	1,190 ± 0.03[b]	1,018 ± 0.06[a]
C17:1	0,326 ± 0.03[c]	0,053 ± 0.07[b]	0,005 ± 0.01[a]
C18:0	8,210 ± 0.09[a]	15,839 ± 0.15[b]	15,886 ± 0.12[b]
C18:1n9c	20,963 ± 0.17[a]	30,043 ± 0.11[b]	19,458 ± 0.09[a]
C18:2n6c	0,183 ± 0.05[a]	0,149 ± 0.07[a]	1,605 ± 0.07[b]
C20:0	0,437 ± 0.04[a]	2,259 ± 0.09[c]	1,034 ± 0.08[b]
C18:3n3	0,192 ± 0.07[a]	0,491 ± 0.01[b]	0,541 ± 0.03[b]
C20:1n9	1,223 ± 0.02[c]	0,082 ± 0.01[a]	0,741 ± 0.07[b]
C21:0	0,000 ± 0.00[a]	0,156 ± 0.09[c]	0,119 ± 0.03[b]
C22:0	0,000 ± 0.00[a]	0,285 ± 0.07[c]	0,225 ± 0.03[b]
C20:3n6	0,090 ± 0.07[a]	0,140 ± 0.05[c]	0,111 ± 0.08[b]
C23:0	0,000 ± 0.00[a]	0,187 ± 0.01[c]	0,091 ± 0.02[b]
C22:2	0,000 ± 0.00[a]	0,110 ± 0.04[b]	0, 074 ± 0.07[a]
C20:5n3	0,000 ± 0.00[a]	0,040 ± 0.01[c]	0,019 ± 0.09[b]
C24:0	0,048 ± 0.03[a]	0,154 ± 0.07[c]	0,117 ± 0.02[b]
C22:6n3	0,074 ± 0.07[b]	0,071 ± 0.07[b]	0,036 ± 0.01[a]

*Means in the same raw followed by different letters represent significant differences($p<0.05$). Data are expressed as mean ± standard deviation.

and 4.17%. In the same study, the amounts of SFA were determined as 72.92% and 67.69% in goat and ewe cheeses, respectively. MUFA and PUFA contents were determined as 23.03% and 4.04% in goat cheese and 26.83% and 5.48% in ewe cheese, respectively.

In our study, the AI value of cow cheese was found to be statistically significant as 3.65, which is higher than that of sheep and goat cheeses ($p<0.05$). TI value was found to be the highest in cow cheese i.e.,4.1405. Milewski *et al* (24). found the lowest AI and highest TI values in sheep and goat cheese samples in a study. In the study of *Aguilar et al.* (25), the opposite result was reported, with the highest AI and lowest TI values in goat and sheep cheese samples. Ulbricht and Southgate suggest that AI and TI indices can characterize atherogenic and thrombogenic potential more strongly

Table 3. Health lipid indices of traditional Kargı Tulum cheese*

	Cow cheese	Sheep cheese	Goat cheese
SFA	73,732 ± 0.95a	66,194 ± 1.05b	75,900 ± 0.97a
UFA	26,269 ± 0.66a	33,805 ± 0.54b	24,098 ± 0.69a
SFA / UFA	2,807 ± 0.00b	1,958 ± 0.00a	3,150 ± 0.00c
MUFA	25,730 ± 0.40a	32,804 ± 0.33b	21,712 ± 0.34a
PUFA	0,539 ± 0.26a	1,001 ± 0.21b	2,386 ± 0.35c
AI	3,650 ± 0.00c	2,023 ± 0.00a	2,931 ± 0.00b
TI	4,1405 ± 0.00c	2,5903 ± 0.00a	3,9824 ± 0.00b
H / H	0,3774 ± 0.00a	0,7899 ± 0.00c	0,5100 ± 0.00b
HPI	0,2739 ± 0.00a	0,3524 ± 0.00a	0,2512 ± 0.00a
DFA	34,479 ± 0.00a	49,644 ± 0.00b	39,984 ± 0.00a
OFA	56,484 ± 0.00c	39,301 ± 0.00a	42,827 ± 0.00b

*Means in the same raw followed by different letters represent significant differences ($p<0.05$). Data are expressed as mean ± standard deviation.

Table 4. Texture Profile Analysis Results of Kargı Tulum Cheese*

	Hardness	Adhesiveness	Springiness	Cohesiveness	Gumminess	Chewiness	Resilience
Cow cheese	3700,871 ± 1564b	-42,042 ± 21.12a	0,777 ± 0.092b	0,0875 ± 0.017a	320,356 ± 125a	251,411 ± 110.11a	0,0275 ± 0.071a
Sheep cheese	1838,757 ± 1423a	-139,239 ± 1.36c	0,687 ± 0.075b	0,1985 ± 0.021a	364,621 ± 230a	257,144 ± 135.54a	0,0505 ± 0.091b
Goat cheese	4007,607 ± 1187c	-91,395 ± 43.23b	0,411 ± 0.037a	0,165 ± 0.071a	659,787 ± 217b	271,101 ± 145.49a	0,0680 ± 0.077b

*Means in the same column followed by different letters represent significant differences($p<0.05$). Data are expressed as mean ± standard deviation.

than dietary PUFA/SFA ratio. The TI and AI indices are related to human nutrition and are also associated with the risk of developing cardiovascular disease. The low AI value in milk and dairy products may provide a protective effect against coronary heart diseases (13). The health indices value of dairy product can be changed regional vegetation of PDO status cheese like Kargı Tulum cheese. The HPI was proposed by Chen et al. (8) as an indicator of the health value of dietary fat. High HPI value of dairy product are assumed to be more beneficial to human health. HPI values ın our study were found to be relatively high. And the highest HPI value of sample was detected in the sheep cheese group (0,35), followed by the cow and goat cheese groups (0,27 and 0,25, respectively).

Table 3 shows that the desired hypocholesterolemic fatty acids (DFA) value of sheep cheese is the highest (49.644). The content in cow and goat cheeses was found to be statistically significantly low ($p<0.05$). The undesirable hypercholesterolemic (OFA) effect of fatty acids in cheeses has also been demonstrated. Milewski *et al* (24), in a study examining the DFA and OFA values in sheep cheeses, found these values as 35.83 and 64.18, while they found theses values as 40.47 and 59.57 in goat cheeses, respectively.

The texture of cheese is an important parameter that affects the purchasing profit and taste of consumers (26). Texture Profile Analysis results of samples are shown in Table 4. The important parameter is the hardness value in tissue profile analysis and most describing the textural properties of the samples. Hardness amounts of samples changed from 1838,757 to 4007,60 g. According to Table 4, the hardness values of cheeses vary over a wide range. The amount of fat,

moisture and mineral contents have important effects on hardness and firmness of cheese (27). The adhesiveness of cheese can be characterized by the moisture. The adhesiveness values of samples ranged between -42,02 and -139,239 g.s. The findings obtained in our study are slightly higher than those of Köse *et al.* (28), for traditional cheese sample, but they were lower than the results from Tarakçı and Deveci(29) for White cheese. Resilience is that it can return to its original shape after the second compression applied to the cheese. Resilience values of samples ranged between 0,0275 and 0,0680 %. Cohesiveness values of cheese samples changed between 0,0875 and 0,1985. These results were different from the study obtained by Köse *et al.* (28), for Malatya cheese. Springiness values of samples changed from 0,411 to 0,777. As can be indicated in Table 4, the highest value obtained for goat cheese sample (659,787) and the lowest for the cow cheese sample (320,356). Similar results were before literature. Chewiness value is a major quality parameter that affects the desirability of the consumer for product (28,30,31). The chewiness values differ between cheese samples. We observed statistically significant differences between cheese samples for all texture parameters measured ($p<0.05$).

Conclusion

This study assessed the fatty acid profile, estimated the lipid quality indices and texture pofiles on geographically indicated Kargı Tulum cheese produced from cow, sheep, and goat milk. Results of our investigation suggest significant differences in fatty acid profiles of cow, sheep and goat cheese. The most favourable fatty acid composition and health lipid indices including index of atherogenicity and thrombogenicity index and HPI indces had prepared from cow, sheep and goat cheese. Kargı Tulum cheese also characterized with the texture profile. Hence, among the evaluated Kargı Tulum cheese samples had the most favorable impact on human health. This could be attributed to several factors including breed, feed, season, geographical origin and production process. The results of our study show that the lipid quality indices differs according to the type of milk used in cheese production. Studies shows that cow, sheep, and goat cheeses have different health effects. In line with the results of our study, we hope that the statistically significant differences in the fatty acid profiles of cheese varieties can be used in future studies to classify cheeses of different geographical origins. In addition, these data can be developed in comparison and classifications to be developed based on the nutritional characteristics of sheep, goat, and cow cheeses.

Acknowledgements: The author is grateful to the Hitit University Scientific Technical Application and Research Center (HUBTUAM) for the analysis.

Conflicts of Interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article.

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