

Detection of milk fat adulteration with margarine and palm oil in kajmak

DOI: 10.15567/mljekarstvo.2025.0203

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Received: 12.09.2024. Accepted: 15.03.2025.

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Abstract

Kajmak is a milk fat-based dairy product that is very popular in the Balkans. However, it is often the subject of adulteration. The aim of this study was to determine the detectable level of adulteration of milk fat in kajmak with margarine and refined palm oil based on iodine value analysis and fatty acid profile by gas chromatographic analysis. In addition, a texture analysis of the adulterated kajmak samples was performed. Six treatments of adulterated kajmak were prepared by mixing fresh kajmak with margarine and refined palm oil in proportions of 5 %, 10 % and 20 % of the total fat in the kajmak. When comparing the fatty acid groups, the detectable level of adulteration for both the added margarine and the palm oil was 10 %. The adulteration of kajmak with 5 % margarine and refined palm oil can be detected by analysing the iodine value as well as by measuring the proportion of trans fatty acids in the case of adulteration with margarine or the C18:3 n-3 in kajmak adulterated with refined palm oil. Larger amounts of added adulterants of 10 % and 20 % can be detected by measuring the proportion of most other fatty acids. The textural properties, in particular firmness and spreadability, were statistically significantly changed ($p < 0.05$) when both margarine and refined palm oil were added, but only in an amount of at least 20 %.

Keywords: milk fat adulteration; kajmak; gas chromatography; spreadability; iodine value

Introduction

Kajmak is a milk fat-based dairy product that is highly valued and popular throughout Serbia for its unique sensory properties. Kajmak is also produced in regions of south-eastern Europe and in some Asian regions such as Turkey, Iran, Afghanistan and India (Pudja et al., 2008; Çakmakçı and Hayaloğlu, 2011; Hashemi et al., 2017; Ningtyas et al., 2017). According to its composition and physico-chemical properties, kajmak can be categorized as somewhere between cheese and butter (Pudja et al., 2008). Kajmak is consumed as a spreadable product immediately after production (fresh kajmak) or after a ripening period (mature kajmak). Spreadability is a very important textural property of kajmak that is expressed both in fresh and especially in mature kajmak (Pudja et al., 2005; Mirecki et al., 2017; Jovanovic, 2022).

The structure of kajmak changes considerably during the ripening process. According to Pudja et al. 2005 the structure of fresh kajmak is not far from the structure of fresh cheese, with similar fat and protein content (40-55 % and 5-10 %, respectively). During ripening, the protein content of kajmak decreases and the fat content increases due to serum separation. According to Pudja et al. (2008), the structure of kajmak fat changes during ripening, similar to the structure of butterfat when agglomerates and milk fat grains form (Walstra, 2006). It is hypothesised that the increase in fat content in kajmak leads to an additional change in the grain structure of the fat, followed by further separation of the whey, resulting in the formation of a uniform butter-like structure (Pudja et al., 2008). The structural changes are accompanied by an improvement in spreadability resulting from the increasing continuity of the fat phase and the decrease in moisture content (Pudja et al., 2008).

Milk and dairy products are among the most frequently adulterated foods worldwide (Ionescu et al., 2023; Everstine et al., 2024). In Serbia, kajmak is often adulterated by replacing the original ingredients with cheaper products such as vegetable oils, margarine, corn flour, cellulose, etc. The adulteration of milk fat in fat-based products such as kajmak, has always been a serious problem, as it is economically profitable to partially replace expensive milk fat with cheaper oils (e.g. palm oil, margarine, etc.) without labeling the product (Rani et al., 2015; Ahmed et al., 2020; Gemechu et al., 2021). The adulterants usually have similar properties to milk fat, but can harm human health, mislead consumers and damage the image of these very popular products (Choudhary and Sharma, 2024). In addition, other illegal ingredients are often added to such products to disguise the adulteration of milk fat.

There is a lot of scientific knowledge about the properties of natural fats, but little information about the physical properties of fat mixtures. To assess the authenticity of milk fat in dairy products, appropriate methods of analysis are required. The determination of the fatty acid composition and triacylglycerol profile by gas chromatography are the most effective analytical methods for the detection of foreign fat in milk and dairy products (Kim et al., 2015; Hajrulai-Musliu et al., 2021). However, these methods require expensive

equipment and professional staff to detect adulteration. Although newer spectroscopy methods such as Raman spectroscopy and, in a certain area, aquaphotomics based on near-infrared spectroscopy (Nedeljkić et al., 2016; Yazgan Karacaglar et al., 2019; Muncan et al., 2021), etc. are available, methods that do not require expensive equipment and chemicals, such as chemical fat constants, e.g. iodine value, can be very helpful in detecting vegetable fat in milk fat (Kahyaoğlu and Çakmakçı, 2016; Dyminska et al., 2017; Salem et al., 2019; Radovanovic et al., 2023).

The aim of this study was to determine the detectable content of added margarine and refined palm oil in adulterated kajmak using gas chromatography (GC) and to compare the results with those of iodine value analysis (IV). Additionally, the influence of adulteration on the textural properties of the kajmak samples was investigated.

Materials and methods

Kajmak sample preparation

Fresh kajmak was bought in a local dairy shop ("Kruna comerc", Belgrade, Serbia) and made from cow's milk using the traditional method. Margarine (hard margarine, produced by the company "Vital", Vrbas, Serbia) and refined palm oil (from Malaysia, imported by "Uvita", Debeljaca, Serbia) were purchased at a local market. Various samples of adulterated kajmak were prepared by mixing fresh kajmak with margarine or refined palm oil in proportions of 5 %, 10 % and 20 %, based on the fat content of the kajmak (Table 1). The original fresh kajmak (KCO) was used as a control sample.

Table 1. Control and experimental kajmak samples

Kajmak sample	Added fat, %	
	Margarine	Refined palm oil
KCO	0	0
KM5	5	0
KM10	10	0
KM20	20	0
KP5	0	5
KP10	0	10
KP20	0	20

KCO - control kajmak without added adulterants; KM5 - kajmak with 5 % margarine; KM10 - kajmak with 10 % margarine; KM20 - kajmak with 20 % margarine; KP5 - kajmak with 5 % refined palm oil; KP10 - kajmak with 10 % refined palm oil; KP20 - kajmak with 20 % refined palm oil.

Kajmak composition

Fat content in the control sample was determined by butyrometric method (FIL-IDF, 1986), total nitrogen content was analyzed by Kjeldahl method (AOAC, 1990) on a Kjeltac System (Tecator 1002, Sweden), and total protein content

was calculated by multiplying it with 6.38. Dry matter was determined by drying method at 102 ± 2 °C (FIL-IDF, 1982). Fat in dry matter and protein in dry matter were calculated. All analyses were carried out at least in triplicate.

Capillary GC analysis of fatty acids

The detection and quantification of fatty acids in kajmak samples was performed by capillary GC according to the method described by Trbović et al. (2013). The total lipids were extracted from the kajmak samples through accelerated solvent extraction (ASE 200, Dionex, Sunnyvale, CA, USA). The fatty acid methyl esters (FAME) were determined with a GC Shimadzu 2010 (Kyoto, Japan). Inlet temperature was 250 °C, while chromatographic separation was carried out using HP-88 (100 m x 0.25 mm x 0.20 µm) column with flow of 1.87 ml/min and nitrogen as a carrier gas. Oven temperature was as follows: 50 °C, 1 min, 13 °C/min to 175 °C, 15 min, 4 °C/min to 215 °C, 10 min, 2 °C/min to 230 °C, 10 min. Detector temperature was 280 °C. The chromatographic peaks in the samples were identified by comparing the relative retention times to the FAME peaks in the Supelco 37 Component FAME mix standard. The mix standard contained butyric acid (C4:0), caproic acid (C6:0), caprylic acid (C8:0), capric acid (C10:0), undecanoic acid (C11:0), lauric acid (C12:0), tridecanoic acid (C13:0), myristic acid (C14:0), myristoleic acid (C14:1), pentadecanoic acid (C15:0), cis-10-pentadecenoic acid (C15:1), palmitic acid (C16:0), palmitoleic acid (C16:1), heptadecanoic acid (C17:0), cis-10-heptadecenoic acid (C17:1), stearic acid (C18:0), elaidic acid (C18:1n9t), oleic acid (C18:1n9c), linolelaidic acid (C18:2n6t), linoleic acid (C18:2n6c), arachidic acid (C20:0), γ-linolenic acid (C18:3n6), cis-11-eicosenoic acid (C20:1), linolenic acid (C18:3n3), heneicosanoic acid (C21:0), cis-11,14-eicosadienoic acid (C20:2), behenic acid (C22:0), cis-8,11,14-eicosatrienoic acid (C20:3n6), erucic acid (C22:1n9), cis-11,14,17-eicosatrienoic acid (C20:3n3), arachidonic acid (C20:4n6), tricosanoic acid (C23:0), cis-13,16-docosadienoic acid (C22:2), lignoceric acid (C24:0), cis-5,8,11,14,17-eicosapentaenoic acid (C20:5n3), nervonic acid (C24:1), and cis-4,7,10,13,16,19-docosahexaenoic acid (C22:6n3) as per certificate of analysis provided by producer. The chromatographic peak areas were corrected by the response factors according to Trbović et al. (2013) calculated by the ratios between the peak area of the individual fatty acid methyl ester and that of the internal standard (heneicosanoic acid methyl ester). The relative quantities were expressed as % (by mass) of total fatty acids. Analyses were performed in triplicate.

Determination of iodine value

The determination of the iodine value (IV) was carried out according to the Wijs method (ISO 3961, 2009), which has been described by Dyminska et al. (2017) and Radovanovic et al. (2023). The preparation of the kajmak samples for IV determination was carried out according to the procedure described in detail by Radovanovic et al. (2023). The prepared kajmak samples were filtered through anhydrous Na_2SO_4

(Merck, Germany). To determine iodine value, the weight of filtered kajmak sample (m) was in the range of 0.5500-0.6000 g. The blank probe was prepared similarly, but without kajmak and the blank value was also determined. The titration was carried out with 0.1 mol/L $\text{Na}_2\text{S}_2\text{O}_3$. V_0 and V_1 were the volumes of $\text{Na}_2\text{S}_2\text{O}_3$ solution used for the titration of the blank probe and the kajmak samples, respectively. The IV value was calculated from the equation (1).

$$\text{IV (g/100g)} = ((V_0 - V_1) \times C \times 12.69) / m \quad (1)$$

Where:

C is the molarity of $\text{Na}_2\text{S}_2\text{O}_3$ solution, (mol/L); 12.69 is the conversion factor

Texture analysis of kajmak samples

All the samples were analyzed for their texture properties: firmness, spreadability, stickiness and adhesiveness. The measurements were carried out using the spreadability test according to the method of Bemer et al. (2016) on a TA.XT plus texture analyzer equipped with the TTC spreadability rig and a 5-kg load cell (Stable Micro Systems Ltd., The United Kingdom). The samples were stored in a refrigerator at 10 °C for 24 hours prior to analysis in order to temper and equilibrate them, and the analysis was performed at 10 ± 1 °C. The lower geometry was filled with a kajmak sample and the surface smoothed. The analysis began by lowering the upper (male) geometry at a speed of 1.0 mm/s, penetrating 2.0 mm into the sample and pushing out the product between the geometries at a 45° angle, after which the upper probe was retracted to the starting position.

Force-time curves were plotted where the maximum force at penetration (the peak value) indicates the firmness of the sample and is expressed in g (Bayarri et al., 2012). The area under the curve in the positive part of the coordinate system, expressed in units of force * time (g.s), represents the total amount of work required to perform the shearing process, i.e. to spread the sample on the surface of the female cone (Brighenti et al., 2008). The force of the maximum negative peak indicates the stickiness of the sample and is expressed in g, while the maximum negative area represents the work of adhesion and is expressed in g.s.

The texture properties (firmness (g); spreadability (g.s); stickiness (g) and adhesiveness (g.s)) were calculated from the obtained graphs using a macro function of the software Texture Exponent (Stable Micro Systems Ltd., United Kingdom). At least five replicates of the analysis were performed per sample.

Statistical analysis

Data were analyzed to investigate the influence of palm oil and margarine addition on the iodine value, fatty acid content and texture properties of kajmak samples by using STATISTICA 9.0 (StatSoft, USA) data analysis software. Mean comparisons of the parameters were performed by LSD-test with significance levels at 0.05.

Results and discussion

Kajmak composition

Kajmak (KC0) had a typical chemical composition for fresh kajmak (Official Gazette of the Republic of Serbia No. 34/2014). The fat and protein contents were 58 % and 6.08 %, respectively. Moreover, the fat content in dry matter was 87.18 %, while the protein content in dry matter was 9.14 %. Due to the addition of margarine and refined palm oil, the total fat content was 61 % in samples KM5 and KP5; 64 % in samples KM10 and KP10, and 70 % in samples KM20 and KP20.

Fatty acid composition

The fatty acid composition of the kajmak samples is shown in Tables 2 and 3. The results of the fatty acid profiles in this study are consistent with previous reports confirming that about 60 % of the total fatty acids in kajmak are saturated fatty acids (SFA), i.e. about 30 % monounsaturated fatty acids (MUFA) (Milićević et al., 2022). The predominant fatty acids were palmitic acid (C16:0) and oleic acid (C18:1 n-9 cis), which is also consistent with the results of Milićević et al. (2022) for kajmak from the Zlatibor area in Serbia.

A comparison of the total amount of SFA showed that the addition of 10 % margarine significantly reduced the SFA content ($p < 0.05$). The reason for this is the difference in the proportion of

SFA in kajmak as a dairy product compared to their proportion in margarine (about 40 %), which is consistent with the results of Vučić et al. (2015), according to which hard margarines in Serbia have a saturated fatty acid content of 35-51 %.

In contrast to margarine, however, the addition of 10 % refined palm oil significantly changed the content of all fatty acid groups (SFA, MUFA, PUFA) in the adulterated kajmak samples, so that the adulterations could be detected at an added amount of 10 % by determining these fatty acid groups (Tables 2 and 3). This is due to the fact that the refined palm oil used contains 45.3 % SFA, which is consistent with the results of Rahman et al. (2022), according to which refined palm oil from Indonesia contains about 50 % SFA, with a high proportion of palmitic acid (C16:0), which accounts for about 44 % of the total fatty acids. In addition, refined palm oil is characterised by a high content of monounsaturated fatty acids, in particular oleic acid, C18:1 n-9 cis (approx. 41 %) and polyunsaturated fatty acid, linoleic acid C18:2 n-6 (8-10.8 %) (Sundram et al., 2003; Gee, 2007; Rahman et al., 2022; El-Nabawy et al., 2023; Lee et al., 2023), which are significantly higher than those of kajmak. Increasing the proportion of added refined palm oil led to a gradual increase in the proportion of these fatty acids (oleic acid and linoleic acid) in the adulterated kajmak samples, which is consistent with the results of El-Nabawy et al. (2023).

The adulteration of kajmak with margarine and refined palm oil can also be determined by comparing the proportion of individual fatty acids. A proportion of 20 % added margarine can be detected by measuring the content of most fatty acids, e.g. saturated with short-chain (C4:0-C12:0) and medium-

Table 2. Fatty acid composition of kajmak samples adulterated with margarine

Fatty acid, %	Kajmak sample			
	KC0	KM5	KM10	KM20
C4:0	4.35±0.08 ^a	4.13±0.10 ^a	3.42±0.10 ^{ab}	2.36±0.07 ^c
C6:0	2.10±0.05 ^a	2.08±0.10 ^a	1.89±0.10 ^{ab}	1.20±0.05 ^b
C8:0	1.00±0.05 ^a	1.05±0.07 ^a	0.97±0.06 ^a	0.60±0.05 ^b
C10:0	1.91±0.06 ^a	1.95±0.09 ^a	1.94±0.07 ^a	1.08±0.05 ^b
C12:0	1.93±0.05 ^a	2.51±0.05 ^b	2.71±0.07 ^{ab}	2.85±0.07 ^c
C14:0	8.24±0.10 ^a	7.79±0.09 ^b	8.05±0.08 ^{ab}	7.80±0.09 ^b
C15:0	0.74±0.05 ^a	0.73±0.05 ^{ab}	0.77±0.05 ^{ab}	0.49±0.06 ^b
C16:0	30.41±0.09 ^a	27.77±0.10 ^{ab}	27.02±0.10 ^{ab}	26.83±0.09 ^b
C16:1	2.13±0.08 ^a	1.81±0.10 ^{ab}	1.37±0.05 ^b	1.21±0.05 ^b
C17:0	0.64±0.05 ^a	0.56±0.07 ^{ab}	0.50±0.05 ^{ab}	0.43±0.05 ^b
C18:0	11.10±0.10 ^a	10.98±0.10 ^{ab}	11.01±0.10 ^{ab}	8.44±0.05 ^c
C18:1 n-9 cis	29.39±0.11 ^a	28.98±0.10 ^{ab}	29.15±0.10 ^{ab}	30.55±0.09 ^b
C18:1 trans-11	0.58±0.05 ^a	3.56±0.06 ^b	5.44±0.05 ^c	8.45±0.08 ^d
C18:2 n-6	2.13±0.06 ^a	2.89±0.10 ^a	3.05±0.05 ^b	5.58±0.05 ^c
C18:3 n-3	0.55±0.05 ^a	0.75±0.09 ^{ab}	0.67±0.08 ^b	0.88±0.09 ^b
MUFA	32.10±0.09 ^a	34.35±0.10 ^{ab}	35.96±0.10 ^{ab}	40.21±0.09 ^{bc}
PUFA	2.68±0.05 ^a	3.64±0.09 ^{ab}	3.72±0.10 ^{ab}	6.46±0.08 ^c
SFA	62.42±0.10 ^a	59.55±0.10 ^{ab}	58.28±0.08 ^b	52.08±0.07 ^c

Results are presented as the mean ± standard deviation. Different lowercase letters indicate significant differences between the kajmak samples within the same row ($p < 0.05$).

KC0 - control kajmak without added adulterants; KM5 - kajmak with 5 % margarine; KM10 - kajmak with 10 % margarine; KM20 - kajmak with 20 % margarine; KP5 - kajmak with 5 % refined palm oil; KP10 - kajmak with 10 % refined palm oil; KP20 - kajmak with 20 % refined palm oil. MUFA (monounsaturated fatty acids); PUFA (polyunsaturated fatty acids); SFA (saturated fatty acids).

chain (C14:0-C17:0) as well as unsaturated fatty acids such as C18:1 n-9 cis; C18:2 n-6; and C18:3 n-3 fatty acids (Table 2).

The exceptions were palmitoleic acid C16:1 and trans fatty acid, vaccenic acid C18:1 trans-11. The addition of 10 % margarine significantly reduced the content of C16:1 ($p < 0.05$). The adulteration of the kajmak fat by the addition of only 5 % margarine (sample KM5) can be demonstrated by determining the proportion of C18:1 trans-11 fatty acid, which was statistically significantly higher compared to the control sample ($p < 0.05$). This result was expected, considering that margarines are known for their high content of trans fatty acids. According to Vučić et al. (2015), very high amounts of trans fatty acids (TFA) were found in hard margarines on Serbian markets (e.g. "Vital" margarine), up to 28.84 % of the total fatty acids.

The adulteration of milk fat by 5 % refined palm oil can be demonstrated by measuring the content of the polyunsaturated fatty acid α -linolenic acid C18:3 n-3 (Table 3). According to Kazazić et al. 2021, butyric acid (C4:0), exclusively present in milk fat (butter), is used to evaluate the lower to middle value of milk fat content in the fats of vegetable and animal origin. In this study, the addition of 10 % palm oil led to a statistically significant ($p < 0.05$) decrease in many saturated fatty acids such as C4:0, C6:0, C12:0, C14:0 and an unsaturated fatty acid C16:1 as well as an increase in unsaturated fatty acids such as C18:1 cis-9 and C18:2 n-6, the content of which can serve as an indicator of adulteration. At the same time, the content of C18:0 and C14:0 may indicate a 20 % adulteration by refined palm oil. Other researchers have come to similar conclusions (Kim et al., 2015; El-Nabawy et

al., 2023). In addition, El-Nabawy et al. (2023) found that a decrease in C14:0 content correlates very well with higher amount of added palm oil to milk fat of more than 20 %. Therefore, it served as a good indicator for adulterating milk fat with palm oil.

Iodine value

The iodine value is a measure of unsaturation in oils and fats. The average iodine value for milk fat is about 32-37 (Salem et al., 2019; El-Nabawy et al., 2023). Although oleic acid accounts for about 30-40 % of all fatty acids represented, milk fat's iodine value is lower than most other fats' iodine value. For this reason, this chemical constant can be used to determine the presence of other fats and oils that contain significantly more or less unsaturated fatty acids.

The milk fat of kajmak has a similar iodine value to butter or milk fat in general (Table 4), which is consistent with the results on the iodine value of kajmak in the literature (Milićević et al., 2022; Radovanovic et al., 2023).

The iodine value for the refined palm oil used was 55.80, which is consistent with literature data for palm oil from Malaysia, which ranges from 50 to 55 (Gee, 2007; El-Nabawy et al., 2023). Due to the high contents of oleic acid C18:1 and linoleic acid C18:2 (Lee et al., 2023), the iodine value of palm oil is significantly higher than the iodine value of kajmak fat (Table 4). Therefore, even a small amount of added refined palm oil (5 %) can be detected by determining the iodine value of adulterated kajmak.

Table 3. Fatty acid composition of kajmak samples adulterated with refined palm oil

Fatty acid, %	Kajmak sample			
	KC0	KP5	KP10	KP20
C4:0	4.35±0.08 ^a	3.79±0.10 ^{ab}	3.18±0.06 ^b	3.01±0.05 ^b
C6:0	2.10±0.05 ^a	1.82±0.10 ^{ab}	1.52±0.05 ^b	1.51±0.06 ^b
C8:0	1.00±0.05 ^a	0.86±0.09 ^{ab}	0.72±0.10 ^{ab}	0.83±0.10 ^{ab}
C10:0	1.91±0.06 ^a	1.63±0.09 ^{ab}	1.33±0.10 ^{ab}	1.37±0.10 ^{ab}
C12:0	1.93±0.05 ^a	1.67±0.08 ^{ab}	1.37±0.06 ^b	1.24±0.05 ^c
C14:0	8.24±0.10 ^a	7.05±0.10 ^{ab}	5.84±0.08 ^c	5.38±0.08 ^c
C15:0	0.74±0.05 ^a	0.65±0.10 ^a	0.56±0.07 ^{ab}	0.52±0.07 ^{ab}
C16:0	30.41±0.09 ^a	31.65±0.08 ^{ab}	31.26±0.09 ^{ab}	33.58±0.09 ^b
C16:1	2.13±0.08 ^a	1.95±0.10 ^{ab}	1.71±0.06 ^b	1.42±0.06 ^c
C17:0	0.64±0.05 ^a	0.59±0.08 ^a	0.53±0.05 ^a	0.48±0.06 ^{ab}
C18:0	11.10±0.10 ^a	10.21±0.09 ^{ab}	10.5±0.08 ^{ab}	8.82±0.07 ^b
C18:1 n-9 cis	29.39±0.11 ^a	31.69±0.10 ^{ab}	33.72±0.08 ^b	34.24±0.10 ^b
C18:1 trans-11	0.58±0.05 ^a	0.5±0.06 ^{ab}	0.62±0.05 ^b	0.60±0.05 ^{ab}
C18:2 n-6	2.13±0.06 ^a	3.12±0.07 ^b	4.16±0.07 ^c	4.28±0.06 ^c
C18:3 n-3	0.55±0.05 ^a	0.28±0.05 ^b	0.25±0.07 ^{bc}	0.23±0.05 ^c
MUFA	32.10±0.09 ^a	34.23±0.10 ^{ab}	36.05±0.08 ^b	36.26±0.06 ^c
PUFA	2.68±0.05 ^a	3.40±0.10 ^{ab}	4.41±0.07 ^c	4.51±0.06 ^d
SFA	62.42±0.10 ^a	59.92±0.09 ^{ab}	56.87±0.06 ^b	57.94±0.08 ^b

Results are presented as the mean ± standard deviation. Different lowercase letters indicate significant differences between the kajmak samples within the same row ($p < 0.05$).

KC0 - control kajmak without added adulterants; KM5 - kajmak with 5 % margarine; KM10 - kajmak with 10 % margarine; KM20 - kajmak with 20 % margarine; KP5 - kajmak with 5 % refined palm oil; KP10 - kajmak with 10 % refined palm oil; KP20 - kajmak with 20 % refined palm oil. MUFA (monounsaturated fatty acids); PUFA (polyunsaturated fatty acids); SFA (saturated fatty acids)

Table 4. The iodine value of kajmak samples

Sample	Iodine value, g/100 g	Sample	Iodine value, g/100 g
KC0	36.56±0.06 ^a	KC0	36.56±0.06 ^a
KM5	41.19±0.66 ^b	KP5	42.08±0.39 ^b
KM10	44.02±0.72 ^c	KP10	43.17±0.65 ^b
KM20	50.90±0.80 ^d	KP20	48.29±0.47 ^c

Results are presented as the mean ± standard deviation. Different lowercase letters (after the standard deviation) indicate significant differences between the kajmak samples within the same column ($p < 0.05$).

KC0 - control kajmak without added adulterants; KM5 - kajmak with 5 % margarine; KM10 - kajmak with 10 % margarine; KM20 - kajmak with 20 % margarine; KP5 - kajmak with 5 % refined palm oil; KP10 - kajmak with 10 % refined palm oil; KP20 - kajmak with 20 % refined palm oil.

According to Vučić et al. (2015), hard margarines produced in Serbia, such as “Vital” margarine, are characterized by high content of trans fatty acids (up to 28 %), MUFA (27-33 %) and PUFA (8-15 %), which leads to high value of iodine value. The iodine value of the Vital margarine used in the experiment was 58.30, which is quite higher than the iodine value of the kajmak control sample (Table 4). Consequently, the iodine value of adulterated kajmak with only 5 % margarine was statistically significantly different ($p < 0.05$) from the iodine value of pure kajmak, which is consistent with the literature data on adulteration of milk fat with margarine (Kahyaoğlu and Çakmakçı, 2016). These authors found that the IV in butter samples adulterated with margarine increased and was proportional to the amount of margarine added (from 0-100 %), whereby the IV of the sample with the addition of 10 % margarine was statistically significantly different from the value of the pure butter sample ($p < 0.01$). Also, according to the results of the iodine value of adulterated butter reported by El-Hasanin et al. (2018), fat constants such as the iodine value can be reliably used to distinguish butter with added margarine from pure butter.

Kajmak texture

Texture is an important attribute for many consumers as it influences the usability and enjoyment of the product. It is known that spreadability as a textural property of cream cheese, margarine, butter and butter substitutes is crucial for consumer acceptance (Ziarno et al., 2023). Kajmak is also characterized by good spreadability and its texture is considered to be between full-fat cream cheese and butter (Pudja et al., 2008).

Brunner (1974) found that differences in the composition of milk fatty acids could explain 80 % of the differences in butter texture. It is known that the fatty acid composition of butter affects its rheological properties, so there are summer and winter butter, which differ in the content of saturated and unsaturated fatty acids and consequently in the iodine value (Youness, 1991; Couvreur et al., 2006; Blaško et al., 2010; Staniewski et al., 2021). This was confirmed by a study by Bobe et al. (2003), which found that butter samples from milk with a higher unsaturated fatty acid content were more spreadable, softer and less sticky.

Butter, as a highly valued product, is often replaced by margarine. It is known that the FA composition of margarine depends on the fat source, production process, etc. Still, a certain amount of unfavorable SFA and TFA is necessary to achieve a firm consistency of margarine (Vučić et al., 2015). Therefore, margarine manufacturers make great efforts to produce hard margarine with texture properties as similar as possible to butter (especially spreadability) and the lowest possible content of trans fatty acids (Silva et al., 2021).

According to DeMan et al. (1990), spreadability is a physical property and results from the fact that full-fat products consist of a dispersion of more or less solid fat crystals in liquid oil. Couvreur et al. (2006) demonstrated that the C16:0/C18:1 ratio significantly influences the melting temperature of milk fat and the sensory perception of properties important to the consumer such as the hardness and spreadability of butter. Summer butter has a lower C16:0/C18:1 content than winter butter (1.32 vs. 1.74), which is associated with a lower butter hardness and better melting behaviour in the mouth

Table 5. Texture of kajmak samples

Kajmak sample	Firmness, g	Spreadability, g.s	Stickiness, g	Adhesiveness, g.s
KC0	513.70 ± 29.02 ^a	543.35 ± 37.23 ^a	-450.78 ± 42.11 ^a	-146.26 ± 5.04 ^a
KM5	484.48 ± 6.51 ^a	505.77 ± 14.88 ^a	-407.87 ± 16.79 ^a	-144.31 ± 13.54 ^a
KM10	536.84 ± 102.95 ^a	556.98 ± 99.32 ^a	-376.90 ± 101.00 ^a	-106.47 ± 19.14 ^a
KM20	1302.52 ± 121.14 ^b	1356.02 ± 7.92 ^b	-1137.99 ± 150.25 ^b	-198.18 ± 26.32 ^b
Kajmak sample	Firmness, g	Spreadability, g.s	Stickiness, g	Adhesiveness, g.s
KC0	513.70 ± 29.02 ^a	543.35 ± 37.23 ^a	-450.78 ± 42.11 ^a	-146.26 ± 5.04 ^a
KP5	818.47 ± 20.75 ^b	810.37 ± 150.45 ^b	-999.48 ± 114.04 ^b	-210.52 ± 28.89 ^b
KP10	917.46 ± 131.39 ^c	982.83 ± 101.25 ^c	-1373.80 ± 176.41 ^c	-194.81 ± 41.76 ^b
KP20	1190.45 ± 98.71 ^d	1524.97 ± 123.35 ^d	-1396.76 ± 13.79 ^c	-297.95 ± 83.50 ^c

Results are presented as the mean ± standard deviation. Different lowercase letters (after the standard deviation) indicate significant differences between the kajmak samples within the same column ($p < 0.05$).

KC0 - control kajmak without added adulterants; KM5 - kajmak with 5 % margarine; KM10 - kajmak with 10 % margarine; KM20 - kajmak with 20 % margarine; KP5 - kajmak with 5 % refined palm oil; KP10 - kajmak with 10 % refined palm oil; KP20 - kajmak with 20 % refined palm oil.

(Blaško et al., 2010). For kajmak, the ratio is around 1.00 (data not shown), suggesting that kajmak is characterised by higher firmness and lower spreadability compared to winter butter.

According to research by Vučić et al. (2015), hard margarines on the Serbian market have a spreadability index of 0.75-1.00, meaning that margarines are similar to kajmak according to this parameter.

However, unlike margarine and palm oil, kajmak contains proteins that, according to Bayarri et al. (2012), help increase firmness and reduce the product's spreadability. In addition, kajmak also contains a much larger amount of water compared to margarine and palm oil, which has an additional effect on the lower spreadability compared to margarine and palm oil (Paduret, 2021). Our measurements showed that the margarine addition in amount of 20 % significantly increased firmness and spreadability compared to control kajmak sample. The reasons for this could be the lower water content (18.1 %) and the higher fat content (81.5 %) of margarine compared to kajmak and probably the condition of the fat in accordance with the results of Bayarri et al. (2012).

In our experiment, the addition of margarine and refined palm oil increased the firmness of kajmak, with the addition of palm oil having a greater effect than that of margarine (Table 5). In contrast to the added palm oil, the addition of 5 % and 10 % of margarine had no significant effect on the firmness of both the kajmak samples ($p > 0.05$). However, larger amounts of both margarine and palm oil (20 %) significantly increased the firmness of the kajmak as well as its spreadability (Table 5).

The results regarding texture could be explained by the fact that kajmak is classified as a product with properties between cheese and butter, additionally it is considered that the texture of fresh kajmak is similar to that of full-fat cheese (Pudja et al., 2008). The reason for this could also be the state of the milk fat at 10 °C, as shown by the results of Bayarri et al. (2012). According to these authors, the proteins in full-fat cheese influence the mechanical properties, i.e. the higher protein content of the cheese influences the structure of the product by increasing the number of bonds between the protein aggregates and improving the strength of the protein matrix and thus of the product. At temperatures as low as 10 °C, the fats become the dominant factor affecting the product's texture, as they are present in the form of a mixture of solid and liquid fats, which further increases the firmness of the protein matrix.

The addition of margarine and refined palm oil increased the adhesiveness and stickiness of the kajmak samples (Table 5). These values have a negative sign as they represent the energy and force required to separate a probe from the sample during the return stroke, i.e. during consumption. It is the work required to overcome the attractive forces between the surface of the food and other surfaces that come into contact with, such as teeth, tongue, and palate (Nishinari et al., 2019).

Compared to the studies on the adhesiveness of different dairy products, the present study is in line with those that found a positive correlation between fat content and adhesiveness of different dairy products (Paduret, 2021; Aleksic et al., 2023) while other authors reported that cheeses with lower fat content had higher adhesive strength

(Ningtyas et al., 2017). According to Paduret (2021) the fat content, in contrast to the protein content in the dry matter, was positively correlated with the product's adhesiveness. Aleksic et al. (2023), compared studies on the stickiness of different dairy products, and found that there is a positive correlation between the fat content and the stickiness of kajmak samples on the Serbian market. The data from our study are consistent with these conclusions, as the stickiness and adhesiveness of the kajmak sample were increased by the addition of fat-based products such as margarine and palm oil. The addition of 20 % margarine as well as just 5 % palm oil statistically significantly increased the stickiness and adhesiveness of kajmak samples KM20 and KP5 (Table 5).

Conclusion

This study showed that a simple and somewhat outdated method such as iodine value analysis can still be used to successfully detect added margarine and refined palm oil in kajmak. Furthermore, this analysis can prove that relatively small amounts (only 5 %) of margarine and refined palm oil have been added to kajmak.

In contrast, modern chromatographic analyzes such as gas chromatography, which can detect the presence of foreign fats and oils based on the fatty acid profile, showed that it was practically possible to claim the presence of at least 10 % added refined palm oil and 20 % margarine. An exception was the analysis of trans fatty acids, whose proportions were statistically significantly ($p < 0.05$) higher in adulterated kajmak with 5 % margarine, as well as the analysis of the reduced α -linolenic acid (C18:3 n-3) content in kajmak adulterated with 5 % refined palm oil.

Acknowledgements

This paper is a result of research within the Contract (number 451-03-65/2024-03/200116) on the implementation and financing of scientific research work in 2024 between the University of Belgrade, Faculty of Agriculture, and the Ministry of Education, Science and Technological Development of the Republic of Serbia.

Detekcija patvorenja mliječne masti s margarinom i palminim uljem u kajmaku, mliječnom proizvodu na bazi masti

Sažetak

Kajmak je mliječni proizvod na bazi masti koji je vrlo popularan na Balkanu. Međutim, često je predmet krivotvorenja. Cilj ovog istraživanja bio je utvrditi detektabilnu razinu krivotvorenja mliječne masti u kajmaku s margarinom i rafiniranim palminim uljem na temelju analize jednog broja i profila masnih kiselina plinskom kromatografskom analizom. Osim toga, izvršena je analiza teksture patvorenih uzoraka kajmaka. Šest tretmana patvorene mliječne masti pripremljeno je miješanjem kajmaka s margarinom i rafiniranim palminim uljem u omjerima 5 %, 10 % i 20 % u odnosu na ukupne masti u svježem kajmaku. Kada se uspoređuju skupine masnih kiselina, detektabilna razina krivotvorenja i za dodani margarin i za palmino ulje bila je 10 %. Patvorenost kajmaka 5 % margarinom i rafiniranim palminim uljem može se otkriti analizom jednog broja kao i mjerenjem udjela transmasnih kiselina u slučaju patvorenja margarinom ili C18:3 n-3 u kajmaku patvorenom rafiniranim palminim uljem. Veće količine ovih dodanih tvari od 10 % i 20 % mogu se otkriti mjerenjem udjela većine drugih masnih kiselina. Svojstva teksture, posebice čvrstoća i mazivost, statistički su značajno promijenjena ($p < 0,05$) kada su dodani margarin i rafinirano palmino ulje, ali samo u količini od najmanje 20 %.

Ključne riječi: patvorenje mliječne masti; kajmak; plinska kromatografija; mazivost; jedni broj

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