



Effect of vegetable coagulant and lamb rennet on physicochemical composition, fatty acid profile and lipid quality indices of a traditional fresh cheese (*Jben*)

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ABSTRACT

Jben is a popular traditional cheese in Algeria and North Africa, usually made from raw ruminant's milk, especially cow. Artisanal production methods are still used in many regions, especially during the abundant milk production season. This study aimed to evaluate and compare the effect of various coagulants (of animal and plant origin) on physicochemical parameters, fatty acids (FAs) profile and lipid quality indices in the traditional fresh cheese "*Jben*". These parameters were analyzed in *Jben* samples made with vegetable rennet (CVR) from *Cynara cardunculus*, which were compared with *Jben* cheeses made using lamb rennet (CLR). The physicochemical characterization showed that pH, titratable acidity, dry matter, fat, and protein contents were 6.19, 92.5 °D, 43.7 g/100 g, 23.4 g/100 g and 15.2 g/100 g cheese, respectively for CVR; and 5.21, 80.3 °D, 50.1 g/100 g, 20.6 g/100 g and 13.8 g/100 g cheese, respectively for CLR. The FAs composition was determined using gas chromatography-mass spectrometry (GC-MS) method. For CLR, FAs comprised 0.09–28.8% saturated fatty acids (SFAs), 0.85–27.68% monounsaturated fatty acids (MUFAs), and 3.22% of polyunsaturated fatty acids (PUFAs); whereas FAs of CVR included 0.22–27.81% SFAs, 1.10–28.16% MUFAs, and 3.07% PUFAs. The predominant FAs identified in all cheeses were C16:0, C18:0 and C18:1(9), with 28.80%, 13.37%, and 27.68% of total FAs, respectively for CLR, and 27.81%, 13.07%, and 28.16%, respectively for CVR. The lowest level of SFAs was observed in CVR, which had the most beneficial health indices with atherogenicity index (2.19), thrombogenicity index (2.91), desirable fatty acid ratio (49.40%) and high hypocholesterolemic/hypercholesterolemic fatty acid ratio (0.72).

Abbreviations: CLR, cheese made with lamb rennet. CVD; cardiovascular disease. CVR, cheese made with vegetable rennet. FA; fatty acid, GC-MS; gas chromatography-mass spectrometry. LCFA, long chain saturated fatty acid; from C18:0 to C24:0. MCFA, medium chain saturated fatty acid; from C12:0 to C17:0. MUFA, mono unsaturated fatty acid. PUFA; poly unsaturated fatty acid. RDA, redundancy analysis. SCFA; short chain saturated fatty acid, from C4:0 to C10:0. SFA; saturated fatty acid. UFA, unsaturated fatty acid.

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1. Introduction

Food processing aims to ensure the preservation of nutrients to defer consumption in space and time and to expand products with a broad range of textures and tastes to meet sensory requirements (Jeantet et al., 2007). Milk is one of those foods that require adequate processing. Its perishability halts the promotion of the dairy sector, because milk begins to deteriorate approximately a few hours after milking. Given its highly perishable nature, it undergoes many treatments to extend its shelf life, which is why rural inhabitants transform milk into several traditional products (Mutwedu et al., 2018). Traditional foods are part of the heritage of each people, and we meet and live recipes, surrounded by an ancestral know-how passed on from generation to generation. Among these foods, traditional cheeses, which represent one of the most important varieties of cheese produced in the world (Fox et al., 2017; Hayaloglu et al., 2002; Irlinger and Mounier, 2009). Traditional cheeses, in particular those produced from raw milk are associated with the territory of origin, eliciting particular characteristics better known as 'Biodiversity Factors'. These factors are due to the landscape, climate, type and plant composition of natural pasture, animal breed and to all those cultural and historical aspects characterizing a specific geographical area (Senoussi et al., 2021, 2022). The combination of all these factors leads to produce a typical cheese, which has the potential to develop into a territorial brand (Papademas and Bintsis, 2017).

In Algeria, local dairy products including cheeses have been disappearing because of the industrialization of the majority of traditional preparation methods for many original products. Also, the rural exodus provoked the demise of ancestral tradition like the traditional manufacture of these products (Medjoudj et al., 2020). Algerian traditional cheeses are divided into different classes including: processed cheeses (*Medghissa*), ripened cheeses (*Bouhazza*), hard cheeses (*Klila*, *Takammart*), and fresh cheeses (*Ighouane*, *Mechouna*, *Jben*) (Leksir et al., 2019). *Jben* is among the most famous traditional varieties, the production method of which remains in use today, and its consumption is increasing due to its pleasant sensory and nutritional properties (Dahou et al., 2015). Although *Jben* is a traditional cheese widely made and enjoyed throughout the country, especially in the east, no scientific data has been published regarding the effect of various coagulants on its characteristics and compositions. Information about its manufacturing technology is still very limited (Senoussi et al., 2022).

Jben is a traditional fresh cheese, made from raw milk of ruminant curdled by coagulating enzyme of vegetable or animal origin after a stage of spontaneous acidification (Hayaloglu, 2017; Medjoudj et al., 2020). Enzymatic coagulation of milk is the critical step in cheesemaking, and the types of milk-clotting enzymes play an important role in this process (Liu et al., 2021). The effect of these milk-clotting enzymes involves the enzyme-mediated cleavage of κ -casein at the peptide bond Phe105-Met106, leading to casein micelles destabilization which induces milk clotting. In addition, milk-clotting enzymes have a larger proteolytic activity towards α_s - and β -caseins at much lower rates than with κ -casein (Fogeiro et al., 2020). As a result, coagulation agents influence micellar aggregation, which is a key element in the defining of curd features and the ultimate gel hardness, which may affect cheese moisture content, texture and flavor (Gomes et al., 2019). Knowing that, the most enzymatic preparations used for cheesemaking have been extracted from the stomachs of ruminants (calf, lamb), but coagulants from microorganisms and plants have also been used at very early dates (Maskey and Shrestha, 2020). For this reason, in addition to cheesemaking conditions, it is essential to understand microbiological, biochemical modifications and fatty acid characteristics during cheese manufacturing in order to produce a product of high hygienic quality (Benyagoub et al., 2016).

Because nutrition and health statuses of dairy product consumers matter, it is important to determine the fatty acid profile of cheese (Kilcawley and Mannion, 2017). Cheese is an essential source of fat in the human diet as it contains a large amount of several fatty acids (FAs). From a nutritional standpoint, various forms of cheese include a high percentage of digestible fat, meaning that the fat globules are smaller. Its digestibility is in the range of 88–94% (Barac et al., 2018). Nevertheless, saturated fatty acids (SFAs), the main group of FAs in cheese, have been linked to cardiovascular diseases, giving cheese a negative nutritional reputation. However, several studies suggest that SFA is one of the factors responsible of cardiovascular disease (CVD), to date there is not any solid scientific evidence to yet showing a causal relationship between dairy products or milk fat and heart diseases (Kanekanian, 2014). In addition, short-chain FAs, which are abundant in cheese, are crucial for promoting human health. For example, butyric acid has been demonstrated to have anti-inflammatory activity and to prevent the evolution of colon and mammary cancers (Gómez-Cortés et al., 2018; Hanuš et al., 2018). According to the type of milk used and the manufacturing process, the composition and content of fatty acid groups (saturated, monounsaturated and polyunsaturated fatty acids) present in the cheese can differ significantly (Markiewicz-Kęszycka et al., 2013; Paszczyk et al., 2020).

In fact, there is very little research on Algerian cheeses, especially *Jben* products, few data available on the biochemical and FA properties of *Jben* and its manufacturing process. The purpose and novelty of this investigation was to examine and compare the effect of various coagulants (of animal and plant origin) on cheese physicochemical parameters, FA profiles and lipid quality indices in the traditional fresh cheese "*Jben*". In addition, this study aimed to determine the influence of principal component parameters on the distribution and concentration of FAs in *Jben* cheeses made using different milk-clotting agents. Due to the importance of cheese in human diet, this comparative study of *Jben* cheese compounds is expected to deepen our understanding about the nutritional value of traditional dairy products.

2. Materials and methods

2.1. Sample collection and transport

The milk samples were taken just after milking; a quantity of milk (4 L) was taken and put into clean coolers. The samples were then brought directly to the laboratory in sterile and refrigerated containers (4 °C) until analysis. These samples were prepared and collected in different localities in eastern Algeria.

2.2. Preparation of clotting agents

For the animal rennet, we have prepared our rennet from lamb rennet cut into strips then shed dried. Drying time is two weeks in hot seasons but can last up to 1–2 months in cold seasons. While for cardoon flowers (*Cynara cardunculus*) in order to obtain optimum coagulant activity and higher extraction yield, cardoon flowers should be harvested at an advanced stage of flowering (dark purple), from fresh flower heads and well developed then kept in a cloth to dry at room temperature and away from direct sunlight, in a well-ventilated place for 10–20 days (Roseiro et al., 2003; Aquilanti et al., 2011).

2.3. Cheesemaking process

In the traditional procedures of *Jben* preparation, raw cow's milk is usually used. The milk is first strained to remove any coarse impurities it may contain, then it is heated in a container to 40 °C for 10 min, then a piece of lamb rennet locally called *Hakka* (about 1g of rennet paste in 4 L of milk for CLR) is put into a porous cloth and the latter is immersed in the milk while it is moderately heated, causing the milk to curdle. After coagulation, the coagulum is drained and poured into fine cloth bags. These bags are then hung up to let the whey drain at room temperature. The length of time the curd is exposed to air depends on the desired consistency of the paste. Generally, the dough obtained is purely lactic, it is often poorly welded and very humid. Once drained, the curd is divided into small pieces and put in glass boxes. For the second type of *Jben* (CVR), we followed the same process but this time the rennet used consisted of cardoon flowers (a quantity of 2 g of dried flower in 4 L of milk). Knowing that, our process of transforming milk into cheese (*Jben*) followed the protocol and technique described by Lahsaoui (2009) (Fig. 1). For each coagulant, two cheese samples (2 replicates) were produced using the same milk batch.

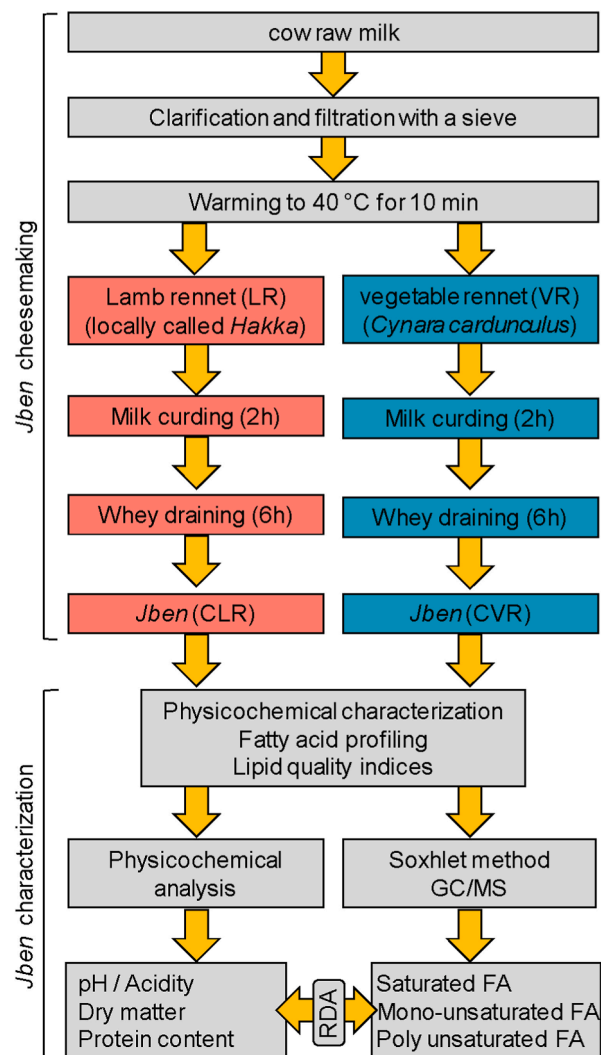


Fig. 1. Diagram of the preparation of the traditional cheese "Jben".

2.4. Determination of physicochemical properties

The following methods were used to determine composition and physicochemical characteristics of cheese samples. All trials were conducted in triplicate.

2.4.1. pH

A slurry was established from a 10 g of ground cheese sample using distilled water (90 mL), then the pH of the homogenate was assessed using a pH-meter (PHM 211, Meter Lab® standard) (Benamara et al., 2016).

2.4.2. Titratable acidity

Total acidity was measured using Nielsen's method. Ten grams of cheese were placed into a 50 mL beaker and then 10 mL of distilled water was added with a graduated pipette and mixed with a magnetic stirrer. The mixture is titrated with a solution of NaOH (0.1N) until the color changed to pink, in the presence of phenolphthalein as a colored indicator (Nielsen, 2017).

2.4.3. Dry matter content

The standard procedure "NF ISO (5534:2004)" was used to measure the content of dry solids of cheese (ISO, 2004). The *Jben* samples were placed in an oven at 102 ± 2 °C for 24 h. Weighing was done after cooling and once the sample has reached a constant weight, the dry extract was calculated.

2.4.4. Total nitrogen content

The ISO standard was used to measure the total nitrogen (8968-1: ISO, 2014). A first step of mineralization was performed on the samples using a K-438 mineralizer coupled to a Scrubber B-414 for 270 mn. Then, the mineral matter was distilled with a mixture of water + sodium hydroxide at 32% and titrated with 0.1N sulfuric acid using a K-370 distillation apparatus (Buchi, Rungis, France), and the protein content was estimated by multiplying total nitrogen by a factor of 6.38.

2.4.5. Fat content

With the use of a Soxhlet extraction equipment, the amount of fat was measured (AOAC, 2005). A weigh of 5 g of grounded and dried cheese wrapped in filter paper were placed in a Soxhlet extraction thimble. Then, a flask filled with a solvent (petroleum ether) was heated so the solvent moves through the sample and extracts the fats and transports them into the flask. The extraction procedure should be continued for nearly 4–5 h. The solvent was evaporated after the extraction was finished, and the amount of lipid remaining was measured.

2.5. Fatty acid analysis

2.5.1. Fat extraction

Gravimetric determination of the fat content (g/100 g of cheese) was carried out after Soxhlet extraction according to the method given in ISO 14156:2001 (ISO, 2001).

2.5.2. Preparation of fatty acid methyl esters

The ISO standard 15884 method was used to convert FA to fatty acid methyl esters (FAMES) (ISO, 2002a). Briefly, in order to dissolve the concentrated fat, 2.5 mL of hexane was mixed with 50 mg of extracted lipid inside an airtight ampoule. A volume of 100 μ L of 2 mol/L methanolic potassium hydroxide was then introduced. Then, 0.25 g of sodium hydrogen sulfate monohydrate (NaHSO₄-H₂O) was added, and the mixture was spun for 3 mn. Finally, GC-MS examination of the produced methyl esters was performed on the top layer.

2.5.3. GC-MS analysis

The IDF technique (15885: ISO, 2002b) was used to evaluate FAMES by GC-MS. The separation of FAMES was quantified by GC (Hewlett Packard Agilent 6890, USA) equipped with Mass Spectrometer (Hewlett Packard Agilent 5973, USA). The conditions for GC-MS analysis were as follows: splitless injection was performed (1 μ L) at 250 °C using an HP-5MS fused silica capillary column with an internal diameter of 0.25 mm and a length of 30 m. The oven temperature was set as follows: 40 °C for 4 min, 10 °C/min, up to 140 °C, isothermal for 1 min, 2 °C/min up to 240 °C, isothermal for 15 min. The analysis took 80 min to complete. At a flow rate of 1 mL/min, Helium (purity N6) was used as the carrier gas. Using a mass detector with the following characteristics: source temperature was maintained at 250 °C, type of ionization was electron impact at 70 eV, analysis mode was scan TIC (30–500 m/z), and mass analyzer type was quadrupole. The identification of common FAs was carried out by comparing their mass spectra data to those acquired using NIST 02.L [US National Institute of Standards and Technology (NIST), Gaithersburg, MD, USA] (Barač et al., 2018).

2.6. Lipid quality indices

To determine the lipid quality indices, the amount of FAs in the fat recovered from cheese samples was employed. They were determined using the subsequent equations.

2.6.1. Index of atherogenicity (IA)

(Equation (1)) (Ulbricht and Southgate, 1991; Omri et al., 2019):

$$IA = \frac{[C12 : 0 + (4 \times C14 : 0) + C16 : 0]}{\sum \text{MUFA} + \sum n-6 + \sum n-3} \quad (1)$$

2.6.2. Index of thrombogenicity (IT)

(Equation (2)) (Willett et al., 1993; Simopoulos, 2002):

$$IT = \frac{(C14 : 0 + C16 : 0 + C18 : 0)}{\left[(0.5 \times \sum \text{MUFA}) + (0.5 \times \sum n-6) + (3 \times \sum n-3) + \left(\frac{\sum n-3}{\sum n-6} \right) \right]} \quad (2)$$

2.6.3. Hypercholesterolemic fatty acids (OFA)

(Equation (3)) (Barac et al., 2018):

$$\text{OFA} = \text{C12:0} + \text{C14:0} + \text{C16:0} \quad (3)$$

2.6.4. Hypocholesterolemic fatty acids (DFA)

(Equation (4)) (Osmari et al., 2011):

$$\text{DFA} = \sum \text{MUFA} + \sum \text{PUFA} + \text{C18:0} \quad (4)$$

2.6.5. Hypocholesterolemic and hypercholesterolemic ratio (H/H)

(Equation (5)) (Ivanova, and Hadzhinikolova, 2015):

$$\text{H/H} = \frac{(\text{C18 : 1n-9} + \text{C18 : 2n-6} + \text{C18 : 3n-3})}{(\text{C12 : 0} + \text{C14 : 0} + \text{C16 : 0})} \quad (5)$$

Where: C12:0 = Lauric acid, C14:0 = Myristic acid, C16:0 = Palmitic acid, C18: 0 = Stearic acid, ΣMUFA = sum of monounsaturated fatty acids, ΣPUFA = sum of polyunsaturated fatty acids, $\Sigma n-3$ = sum of n-3PUFA, $\Sigma n-6$ = sum of n-6PUFA.

2.7. Statistical analysis

Using sample-based data, the variation of each physicochemical parameter (pH, titratable acidity, dry matter content, fatty matter, total nitrogen content and protein content) between CVR and CLR was tested using the non-parametric Kruskal-Wallis one-way analysis of variance (ANOVA). Prior ANOVA testing, normality was verified using Shapiro-Wilk test. Pearson correlations were applied to test the relationships between physicochemical parameters analyzed in *Jben* cheese made using each of the two milk-clotting agents (lamb rennet and flowers of *Cynara cardunculus*). The correlation matrices were visualized in an interactive plots using the package {corrplot} (Wei and Simko, 2021) under R software version 4.2.1 (R Core Team, 2022). Pearson's Chi-squared tests were used to test the variation FA abundances between the CVR and CLR. Statistical significance was fixed to alpha = 0.05. The similarity of FA composition among CVR and CLR was analyzed using two abundance based similarity metrics, namely Morisita-Horn overlap index and Bray-Curtis metric. To understand the relationships of physicochemical characteristics of CVR and CLR on composition and abundance of different FAs, a redundancy analysis (RDA) was executed using the {vegan} package in R.

3. Results

3.1. Physicochemical composition

Fig. 2 illustrates the effect of milk-coagulant type (i.e. lamb rennet and cardoon flowers) on the gross physicochemical composition (pH, titratable acidity, dry matter, fat and protein content) of traditional fresh cheese '*Jben*' made from cows' milk. Most chemical parameters, including pH, titratable acidity and fat content, were quite among the two types of coagulants. However, in all cases, except the protein content, values of the physicochemical parameters differed significantly ($p < 0.05$) between CLR and CVR.

Results of pH indicated that the samples had acidic pH with 5.21 ± 0.35 (mean \pm standard deviation) for CLR and 6.19 ± 0.23 for CVR. The values of titratable acidity and fat content of the samples analyzed averaged 80.3 ± 8.13 °D to 92.5 ± 2.60 °D and 20.6 ± 2.23 g/100 g to 23.4 ± 2.23 g/100 g for CLR and CVR, respectively (Fig. 2). In addition, the lowest dry matter values were determined in CVR (43.7 ± 3.93 g/100 g) whereas CLR showed the highest dry matter content (50.1 ± 7.25 g/100 g). The average levels in proteins were 13.8 ± 2.02 g/100 g cheese for CLR, whereas CVR samples recorded 15.2 ± 2.11 g/100 g.

3.2. Interrelationships between cheese physicochemical parameters

Pearson correlations (Fig. 3) revealed that the fat and protein contents in *Jben* CLR were non-correlated to all other cheese variables. Conversely, there was a good relationship between pH, titratable acidity and dry solids content in LRC. A strongly negative correlation ($r = -0.95$, $p < 0.001$) was found between pH and titratable acidity. A significant negative correlation between dry solids and pH was also observed. On the other hand, a strong positive correlation ($p < 0.05$) was detected between fat and protein contents also between dry matter content and titratable acidity ($r = 0.70$). In regards to CVR, there were many significant correlations among physicochemical parameters (Fig. 3). Strong positive correlations ($p < 0.05$) were found between fat content, protein and dry solids, whereas, the pH was negatively correlated with titratable acidity.

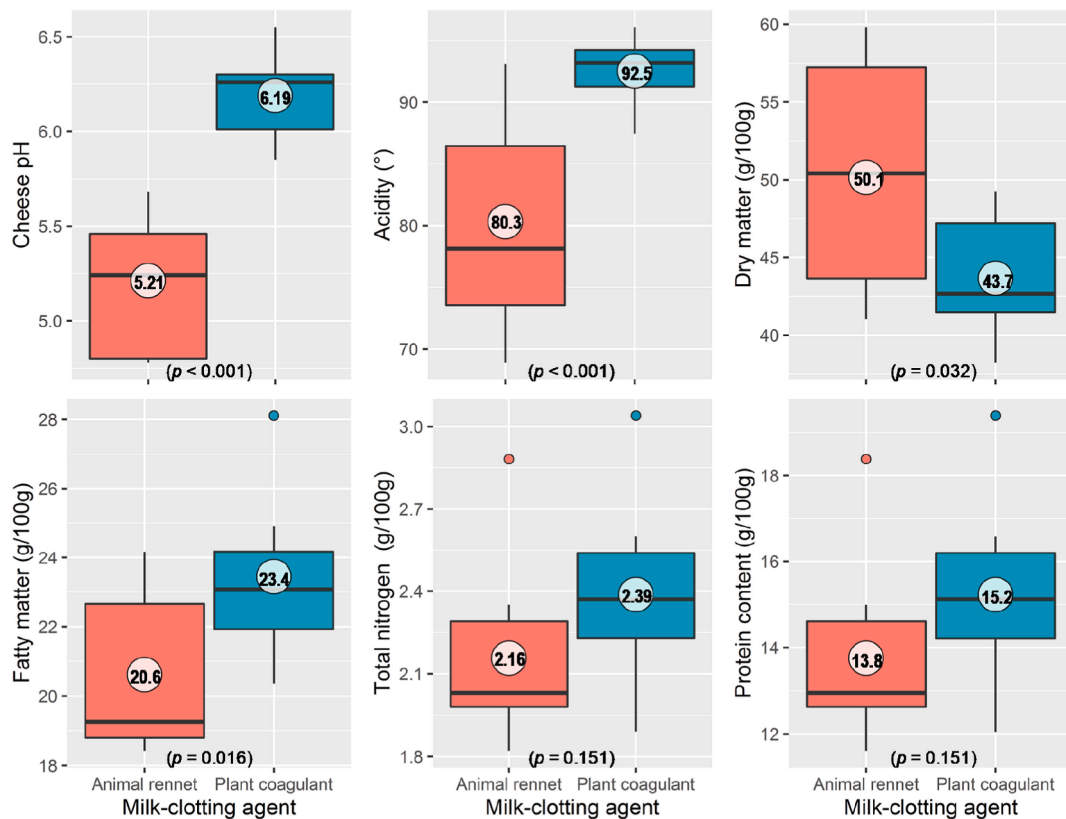


Fig. 2. Variation of principal physicochemical component parameters of *Jben* cheese made using two milk-clotting agents (lamb and vegetable). Figures displayed inside the solid white circles are means. *P*-value presented between brackets within plots are results of Kruskal-Wallis one-way analysis of variance.

3.3. Cheese FAs composition

The composition of FAs in the total fat (% of total FAs) obtained from Algerian traditional fresh cow's cheeses is shown in Table 1. Chromatograms of GC/MS for CLR and CVR samples are given in Fig. 4. According on the type of cheese, up to fifteen FAs were found under the experimental circumstances utilized in this work. Ten of these were SFAs, while the remaining five were UFAs (MUFAs and PUFAs). The studied traditional fresh cow's cheeses generally had identical qualitative fatty acid compositions in both cheeses (Pearson's Chi-squared test: $\chi^2 = 0.361$, $df = 14$, $p = 0.99$). Our finding demonstrated that SFAs were predominant in both cheese samples, constituting 65.88% of FAs in CVR and 64.3% in CLR. MUFAs and PUFAs reached a value of 30.22% and 3.22% for CVR and of 32.63 and 3.07% for CLR, respectively (Fig. 4). The most abundant SFAs in all cheeses were, myristic acid (C14:0), the stearic acid (C18:0) and the palmitic acid (C16:0). These FAs constituted 12.17–11.69%, 13.37–13.07% and 28.80–27.81% of all identified FAs of CVR and CLR, respectively (Fig. 5). Pearson's Chi-squared tests revealed no significant difference ($p > 0.05$) for each FA concentration between CVR and CLR.

The fatty acid that contributed most to the profile of UFAs was oleic acid (C18:1 n9cis). The second most abundant MUFA was C16:1, identified in CVR, which was almost equal to its content CLR. For C17:1 its value in CVR likewise surpassed its amount in CLR. Content of C14:1 was almost as low as the C16:1 content for the highest type (CVR) and for the lowest type (CLR). Oleic acid (C18:1 n-9 cis) found in all of the cheese samples examined correspond to the predominant acid of the MUFA group (Table 1), which values obtained were 28.16 and 27.68 for CLR and CVR, respectively. Regarding PUFAs in CVR and CLR products, only linoleic acid (C18:2 n-6 cis) was present with 3.22% and 3.07%, respectively. In case of short chain saturated fatty acid (SCFA) content, there were no appreciable differences between CLR (5.66%) and CVR (5.46%). The medium chain saturated fatty acid (MCFA) levels in the examined cheeses were comparable. For instance, palmitic acid (C16), which was more prevalent and higher in CLR than CVR, was the most prevalent MCFA, in contrast, myristic acid (C14), which was more abundant in CLR than CVR, was the second most prevalent fatty compound. Though C18 (stearic, oleic and linoleic) was the most representative FA among long-chain fatty acids (LCFAs), this class of FAs was practically the same in CVR (44.52%) as compared with CLR (44.36%).

3.4. Similarity analysis

As shown in Fig. 6, the 15 FAs identified were shared between the two *Jben* cheeses made using two milk-clotting agents. Overall, a total of 15 FAs were determined using GC-MS method. 11l these FAs were present in both CLR and CVR, thus the qualitative similarity reached 100%. However, the abundance based similarity metrics based on these 15 FAs was slightly different but remained very high, with a Bray-Curtis metric = 92% and Morisita-Horn overlap index = 96%.

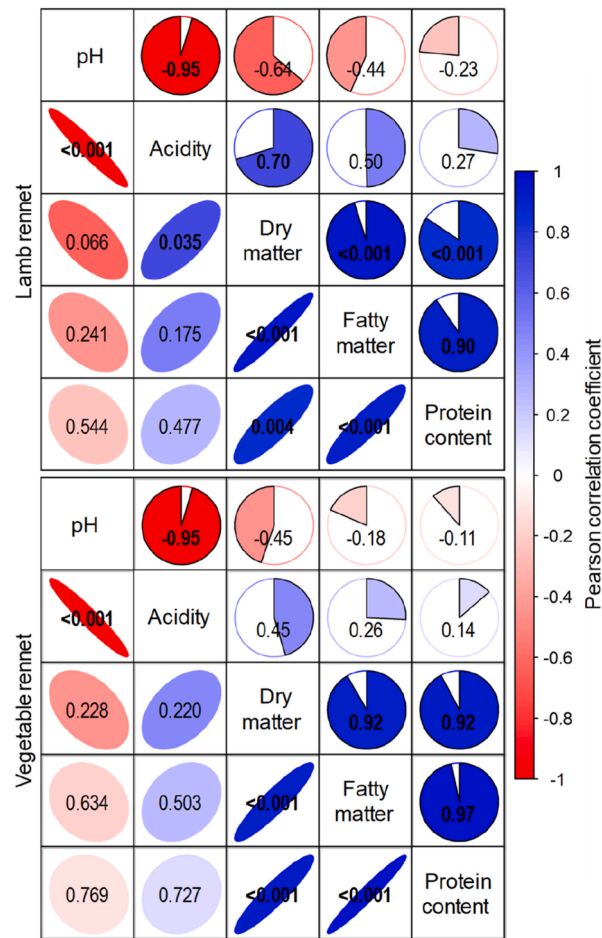


Fig. 3. Correlation matrix between physicochemical parameters analyzed in *Jben* cheese made using two milk-clotting agents (lamb and vegetable). Pearson coefficients are mapped proportionally to values in pie chart and color intensity, whereas p -values of the tests are shown in values below the diagonal.

3.5. Lipid quality indices

The results of the current study indicated that CVR and CLR had slightly different values for the lipid quality indices (Table 2). For the CLR, it was determined that the levels of DFA (47.49), AI (2.43), TI (3.18), and USFA/SFA ratio (0.51) were undesirable. In contrast, the beneficial lipid indices were observed in CVR with DFA (49.4), AI (2.19), TI (2.91) and USFA/SFA (0.55). The CLR and CVR products showed H/H ratios of 0.68 and 0.72, respectively. While, the undesired hypercholesterolemic fatty acids (OFA) value in CLR was lower (43.28) than in CVR (44.88).

In addition, among the health indices investigated, the MUFA/SFA and PUFA/SFA ratios were influenced by the milk-clotting agents. This was due to the higher MUFA content and the lower SFA content of CVR and the lower MUFA and the higher SFA of CLR. MUFA/SFA ratio was higher in CVR (0.50) than in CLR (0.45). Regarding the values for PUFA/SFA found in our cheese samples were at a similar level (0.048 for CLR and 0.048 for CVR). Regarding the CLR, it was determined that the levels of DFA (47.49), AI (2.43), TI (3.18), and USFA/SFA ratio (0.51) were unfavorable.

Additionally, the milk-clotting agents had an impact on the MUFA/SFA and PUFA/SFA ratios among the health indices examined. Due to the lower SFA and higher MUFA levels of CVR, MUFA/SFA ratio was higher in CVR (0.50) than in CLR (0.45). Regarding the PUFA/SFA ratio that we obtained in the analyzed cheese samples, they were at a comparable level (0.048 for CLR and 0.048 for CVR).

3.6. Relationships between physicochemical parameters and fatty acids in both cheeses

The redundancy analysis (RDA) revealed the relationships between physicochemical characteristics and FAs in both cheeses CLR and CVR (Fig. 7). The contribution of each studied variable in the RDA triplot was assessed by the length of each vector. The first factorial axis (RDA1) of the RDA triplot explained 95.3%, and the second factorial axis (RDA2) explained 4.7% of the total variation in the data. The RDA demonstrated that pH, titratable acidity, protein and fat content were positively correlated with FAs from CVR (myristic, pentadecanoic, palmitic, heptadecanoic, margaric oleic and arachidic) suggesting that pH and titratable acidity had strong effects on the FAs in CVR. In contrast, there was a negative association between these FAs and dry matter. Whereas, the FAs: palmitoleic, caproic, caprylic, capric, lauric, myristoleic, stearic and linoleic from CLR were negatively correlated with pH, titratable acid-

Table 1
Fatty acid content of traditional cheeses (% of total fatty acids).

Fatty acid methyl esters		% (CLR)	% (CVR)
Common name	Formula		
Caproic	C6:0	1.52	1.38
Caprylic	C8:0	1.25	1.22
Capric	C10:0	2.89	2.88
Lauric	C12:0	3.91	3.78
Myristic	C14:0	12.17	11.69
Pentadecanoic	C15:0	1.28	1.50
Palmitic	C16:0	28.80	27.81
Margaric	C17:0	0.60	0.75
Stearic	C18:0	13.37	13.07
Arachidic	C20:0	0.09	0.22
Saturated fatty acids (SFAs)		65.88	64.3
Myristoleic	C14:1	0.85	1.10
Palmitoleic	C16:1	1.48	2.21
Heptadecenoic	C17:1	0.89	1.16
Oleic	C18:1 n-9 cis	27.68	28.16
Monounsaturated fatty acids (MUFAs)		30.09	32.63
Linoleic	C18: 2 n-6 cis	3.22	3.07
Poly unsaturated fatty (PUFAs)		3.22	3.07
Unsaturated fatty acids (UFAs)		34.12	35.7
SCFAs		5.66	5.48
MCFAs		49.98	50
LCFAs		44.36	44.52

SFA: Saturated Fatty Acids; MUFA: Mono Unsaturated Fatty Acids; PUFA: Poly Unsaturated Fatty Acids.

SCFAs: Short Chain Saturated Fatty Acids from C4:0 to C10:0; MCFAs: Medium Chain Saturated Fatty Acids from C12:0 to C17:0; LCFAs: Long Chain Saturated Fatty Acids from C18:0 to C24:0.

ity, protein and fat content, but were positively correlated with dry matter. In this study, CLR was characterized by considerable dry matter content and was different from CVR; poor in terms of nutrition status. Because it contained the lowest levels of protein, fat and titratable acidity compared to the other tested cheese. On the contrary, CVR was characterized by its high content of proteins, fat content, titratable acidity and pH.

4. Discussion

4.1. Physicochemical composition

Obviously, the type of rennet utilized in *Jben* cheesemaking had an impact on all parameters examined, including pH, titratable acidity, fat content, dry solids, and protein content levels. Due to the lack of production standards, cheese pH ranged from 5.21 for the CLR to 6.19 for the CVR, with significant variations between samples. ANOVA testing the variation of pH values (Fig. 2) showed that there was significant difference ($p < 0.05$) between (CVR) and (CLR). Yet, these pH measurements in cheese samples are consistent with previous studies (El-Kholy, 2015; Gulzar et al., 2019; Islam et al., 2021). However, the possible variation may be due to the processing techniques, effect of coagulants, incubation and coagulation time and milk-crude temperature. pH of the cheese may vary due to the strength of coagulants (Koirala, 2021). In addition, renneting with the lamb rennet decreased the pH as compared with vegetable rennet, which could be related to a greater activity of lactic acid bacteria. On the other hand, artisan and industrial animal rennet pastes demonstrated a significant microbial diversity, which proved that animal rennet pastes are sources of lactic acid bacterial strains with technologically advantageous characteristics for cheese production (Cruciata et al., 2014; Kajak-Siemaszko et al., 2022).

As shown in Figure (2), it could be noticed that all cheeses were found to have significantly high titratable acidity. The findings of the titratable acidity obtained in this investigation were consistent with previous studies (El-Kholy, 2015; Benyahia et al., 2021). Our results, however, differed from those of previous studies on Algerian cheeses (e.g. Tadjine et al., 2020). A higher lactic acid bacteria activity during cheesemaking may be the reason of these variances between cheeses (CVR and CLR) (Czyzak-Runowska et al., 2020). The fresh CVR and CLR had a total dry extract of 43.7 and 50.1 g/100 g, respectively. Compared with literature data, we found that these averages were lower than those reported by Benyagoub et al. (2016), which was estimated at 56.12%, but it is close to that of Santillo et al. (2007) with 43.1% based on the lamb rennet and enzymatic extract prepared from *C. cardunculus* by El-Galeel et al. (2017) (51.88%). The possible variation in the total solids content between CVR and CLR may be due to curd types obtained, which are dependent on the initial milk composition, effect of coagulants as well as the type of drainage (Benyahia et al., 2021).

Regarding our findings of total fat content, a significant difference ($p < 0.05$) was observed among CLR and CVR, but both had high fat contents. Similar values were revealed by Hashim et al. (2011) and Benyahia et al. (2021). Tofalo et al. (2015) showed significant differences in fat depending on the types of rennet used. On another side, the mean content of protein for studied cheeses did not significantly differ ($p < 0.05$). The average protein content of these cheeses, according to García et al. (2012), ranges between 14.62 and 15.05 g/100 g cheese, which is comparable to the findings of the current study. The general trend of the current findings is consistent with those reported for fresh cow's cheese (Benheddi and Hellal, 2019), fresh ewe's cheese (Gutiérrez-Peña et al., 2021), and fresh

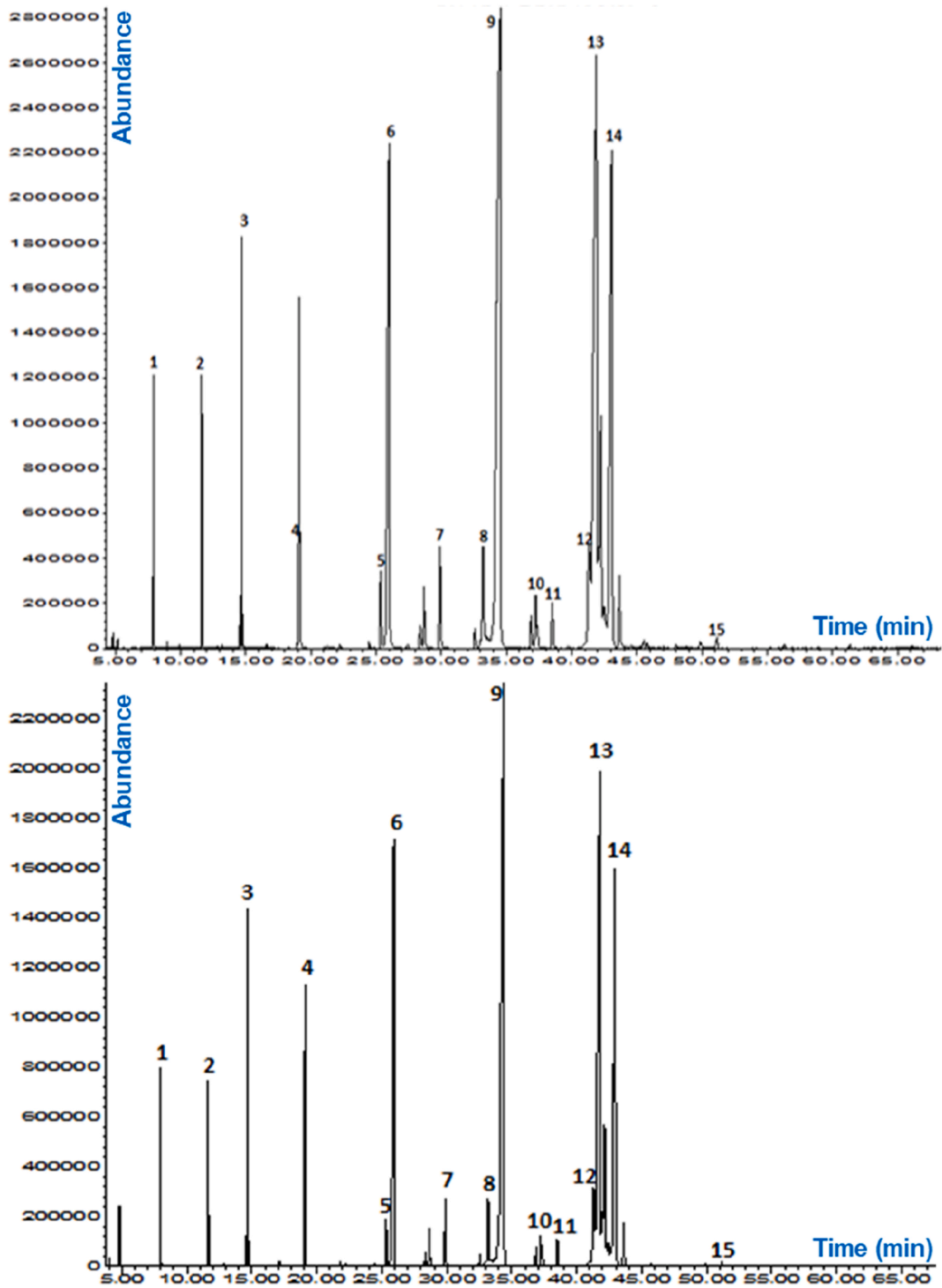


Fig. 4. Chromatogram of separation obtained of sample cheese made with vegetable rennet (upper graph) and of sample cheese made with lamb rennet (bottom graph) products. Figures of the curve peaks are FAs (1: Caproic, 2: Caprylic, 3: Capric, 4: Lauric, 5: Myristic, 6: Pentadecanoic, 7: Palmitic, 8: Margaric, 9: Stearic, 10: Arachidic, 11: Myristoleic, 12: Palmitoleic, 13: Heptadecenoic, 14: Oleic, and 15: Linoleic).

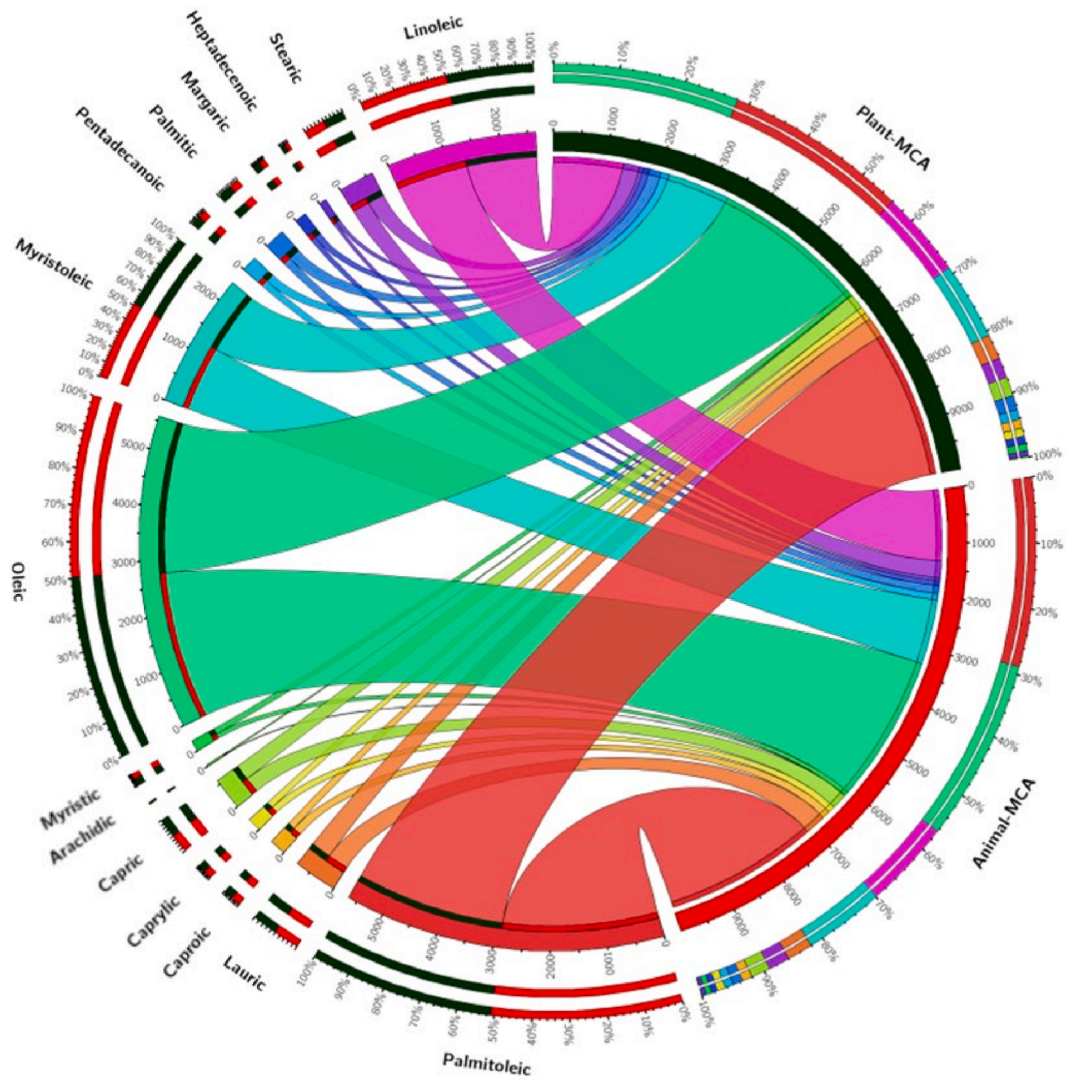


Fig. 5. Chord diagram of fatty acid composition in fat extracted from cheeses made with lamb rennet and vegetable rennet (% of total FAs).

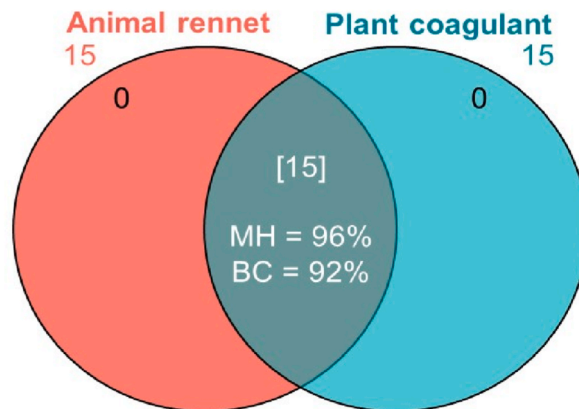


Fig. 6. Venn diagram showing the similarity of fatty acid composition in *Jben* cheeses made using two milk-clotting agents. MH: Morisita-Horn overlap index and BC: Bray-Curtis index are the abundance based similarity metrics.

Table 2
Lipid quality indices of *Jben* cheese prepared using lamb rennet and vegetable rennet based on *Cynara cardunculus*.

Indices	CLR	CVR
Atherogenicity index (IA)	2.43	2.19
Thrombogenicity index (IT)	3.18	2.91
Hypocholesterolemic fatty acids (DFA)	47.49	49.4
Hypercholesterolemic fatty acids (OFA)	44.88	43.28
Hypocholesterolemic/Hypercholesterolemic ratio (H/H)	0.68	0.72
Mono Unsaturated Fatty Acids/Saturated Fatty Acids (MUFAs/SFAs)	0.45	0.50
Poly Unsaturated Fatty Acids/Saturated Fatty Acids (PUFAs/SFAs)	0.048	0.047
Unsaturated Fatty Acids/Saturated Fatty Acids (UFAs/SFAs)	0.51	0.55

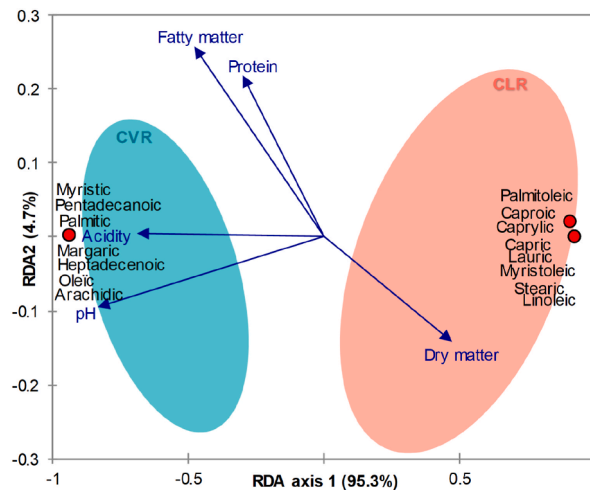


Fig. 7. Triplot of the Redundancy analysis (RDA) exploring the relationships between fatty acids and physicochemical properties of CLR and CVR cheeses.

goat's cheese (Mladenović et al., 2022). Several variables, including cheesemaking practices, milk chemical and microbiological characteristics, animal feeding systems, origin of milk-clotting agents have a strong impact on the content and quality of the cheese (De Marchi et al., 2008; Formaggioni et al., 2015; Uzun et al., 2020).

Based on the above results, Fig. 3 shows the findings of various physicochemical characteristics (pH, acidity, fat, dry solids and protein content) and the relationships between those parameters. For instance, it was found that the pH and acidity values in both cheeses correlated negatively ($p < 0.05$); The action of lactic acid bacteria and the generation of lactic acid are responsible for the pH drop. These findings are consistent with the observations of Ioannidou et al. (2022). Additionally, pH and dry matter showed negative relationships. Both Manca et al. (2020) and de la Haba Ruiz et al. (2016) reported similar findings. Fat and dry matter values in cheeses were found to be strongly and positively correlated. Since the dryness of cheese (CVR) influences the quantity of fat because as moisture declines, fat grows, the positive link between cheese dry solids and fat content seems logical (Ioannidou et al., 2022).

4.2. Fatty acid profile

Besides of being abundant providers of vital nutrients like protein, calcium, and vitamins, milk and dairy products also include a variety of bioactive compounds, of which FAs are of the greatest significance. Dairy fat is the most complex fat in the human diet since it contains more than 400 different FAs. Furthermore, a substantial portion of these FAs comes from dairy products, which constitute almost their entire source in our diets (Summer et al., 2017). Many studies evaluated the profiles of FAs in traditional cheeses (Barać et al., 2018; Çetinkaya and Oz, 2018; Paszczyk et al., 2022). The amount of FAs in dairy products (cream, yogurt, cheese) is related to cheesemaking process and maturation time (Białek et al., 2020; Paszczyk et al., 2020; Uzun et al., 2020; Buccioni et al., 2022).

As expected, SFAs and UFAs are abundant in fresh cheeses, as well as other varieties of cheese. In the FA profiles, SFAs were characterized by the highest proportion in all fresh cheese samples (CVR and CLR). The values for SFAs found in the present study are consistent with those found by Dopieralska et al. (2020). The contents of SFAs, MUFAs, and PUFAs in the studied fresh cheeses (CVR and CLR) made with cow's milk varied from 64.63 to 65.53%, 29.28–30.92%, and 4.27–5.19%, respectively. These results are in line with the data reported by Barać et al. (2018), which showed that the concentration of SFAs, MUFAs, and PUFAs in cheese (Sjenica) derived from cow's milk was 65.97%, 30.31%, and 3.72%, respectively. Moreover, C16:0 (palmitic), C18:1n-9 (oleic) and C18:2.n-6 (linoleic) acids were the main SFA, MUFA and PUFA for Cheeses (CVR and CLR). Paszczyk et al. (2022) also gathered comparable data regarding the previous FA categories. In addition to lauric, palmitic, and palmitic acids, which have been shown to be the most important SFAs, CVR and CLR contain additional FAs that have favorable effects for chronic disease onset and progression, including many odd-chain FAs (OCFAs) and C18:1n-9 (oleic acid). Elevated plasma OCFAs have been connected to increased dairy consumption, particularly C15:0 and C17:0 which can be used as a biomarker of total dairy fat absorption. Higher OCFAs plasma levels have been shown to

be linked with a reduced CVD risk. The studied fresh cheeses had adequate levels of C15:0 and C17:0, particularly in CVR, which had an improved FA profile. The amounts of MUFAs in cheeses (CVR and CLR) were slightly different (Table 1). Oleic acid (C18:1n-9) was the most prevalent acid in the MUFA category in all cheese samples examined. According to Hanuš et al. (2018), MUFAs have been demonstrated to have anti-cancer and anti-atherosclerotic characteristics, so it can be used in daily nutrition. On the other hand, in all cheese samples (CVR and CLR), the proportion of PUFA was the lowest in the total FA group. To prevent certain diseases (cancer, diabetes and CVDs) and benefit human health, sufficient intakes of n-6 and n-3 FAs are necessary (De Lorgeril et al., 1994; Holub and Holub, 2004; Willett, 2007; Simopoulos, 2008). The only n-6 FA found in any of the samples examined, among PUFA, was linoleic acid (C18:2 n-6). Inflammatory, cancerous and CVDs are only just some of the illnesses whose pathogenesis is facilitated by an increased in n-6/n-3 ratio resulting from an excess of n-6 PUFAs. Conversely, higher concentrations of n-3 PUFAs have preventive impacts. According to several research, the ideal ratio may change based on the type of the disease (Simopoulos, 2006; Russo, 2009). In CVR and CLR cheeses, content of linoleic acid varied slightly (3.22% and 3.07%, respectively) (Table 1). As compared with previous data, these values are consistent with those revealed by Paszczyk et al. (2022). Linoleic acid is necessary for the production of arachidonic acid, which serves as a precursor to a wide range of vital compounds such as prostaglandins (essential in inflammation, smooth muscle contraction and blood clots) and leukotrienes (essential in allergic diseases) in ruminant milk (Calder, 2012).

On the other hand, SCFAs play a crucial role in advancing human health. For example, the capric acid (C10:0), which has antiviral and antibacterial effects, is the most prevalent SCFA in the fats of both cheeses (Nair et al., 2005). Nevertheless, despite their very low concentrations in our cheeses, SCFAs are usually responsible for giving cheeses its distinctive flavors (Martino et al., 2019). In contrast, palmitic acid (C16:0) was the MCFA groups predominate FAs in all cheese varieties cheese (Table 1). Similar findings were found by Milićević et al. (2022). The distribution of FA chains, including SCFA, MCFA, and LCFA, differs significantly from that found by Falchero et al. (2010); in particular, MCFA and LCFA contents were 44.16% and 45.73%, respectively, while the mean values of CLR discovered in our research were 49.98% and 44.36%. The slight variation of FAs between CVR and CLR may be attributed to the lipolytic activity resulting from both the starter microorganisms and the rennet paste added to milk during the cheesemaking process (Gutiérrez-Peña et al., 2021).

The RDA was used to assess inter-links between FAs in CLR and CVR and their physiochemical properties (Fig. 7). The projection of FAs and all cheese characteristics on the two first dimensions of RDA triplot stressed that FAs of CVR (myristic, pentadecanoic, palmitic, margaric, heptadecenoic, oleic, and arachidic) had strong positive correlations with acidity, pH, protein and fatty matter contents, because the first RDA axis expressed 95.3% of the explained variance. Similarly, FAs from CLR (capric, caproic, caprylic, lauric, linoleic, myristoleic, palmitoleic, and stearic) were associated to dry matter content. These findings are consistent with previous studies (Abbas et al., 2021; Esmailzadeh et al., 2021), which showed that the oleic was strongly correlated with pH. Moreover, the increase in the dry matter is accompanied by an increase in the fat content (Ioannidou et al., 2022). This study indicated that the effects fat and protein contents are secondary compared to cheese acidity, which is in agreement with the results found by Giosuè et al. (2022).

4.3. Lipid quality indices

Collecting information on lipid quality indices can contribute to a better understanding of the nutritional and/or health effects of dairy products consumption. Atherogenicity (IA), thrombogenicity (IT), desirable fatty acids ratio (DFA), UFA/SFA, MUFA/SFA, and PUFA/SFA ratios data were compared between CLR and CVR in the current study (Table 2). In regard to our findings, cheese types (CLR and CVR) did not showed any significant impact on IA or IT indices, as these were practically the same in CVR as compared with CLR.

The ability to induce platelet aggregation is assessed by the IA and IT indices. The IT defines PUFAs (n-6 and n-3) and MUFAs as antithrombogenic, and SFAs (particularly myristic, palmitic and stearic acids) as thrombogenic (Ulbricht and Southgate, 1991). Lower levels of these indices are known to be useful in preventing blood vessel diseases. In the current study, IA and IT levels in all cheese samples ranged from 2.19 in CVR to 2.43 in CLR and from 2.91 in CVR to 3.18 in CLR, respectively. According to Chen and Liu (2020), there are no recommended values for IT and IA indices. In our work, these levels are consistent with those revealed by Hirigoyen et al. (2018).

Regarding nutritional value, higher levels of H/H ratio have been noted as healthier and to prevent certain diseases such as CVDs. The obtained H/H values for both cheeses were 0.68 and 0.72, respectively, which are in line with those found in other studies (Silva and Bessa, 2002; Paszczyk and Łuczyńska, 2020). Additionally, our research indicates that CVR contains the least undesirable hypercholesterolemic fatty acids (OFA) and most beneficial hypocholesterolemic fatty acids (DFAs). The DFA values in the present study are in agreement with those of Barać et al. (2018). The UFA/SFA ratio was different among *Jben* cheeses (CVR and CLR), these results varied between 0.45 and 0.50, respectively, while it is close to those cited by Barać et al. (2018) for *Sjenica* (0.51) and *Zlatar* (0.46) traditional Serbian cow cheeses and by Çetinkaya and Oz (2018). In order to prevent consumer's health issues, the most prevalent indicator used to assess the nutritional quality of foods including cheese is PUFA/SFA ratio. A higher PUFA/SFA ratio is directly related to a higher value of PUFA, which has beneficial incidence on human health, as they lower blood cholesterol levels, which prevents some contemporary diseases such as cancer and CVDs (Michikawa, 2003; Wood et al., 2004). Values of PUFA/SFA ration reported in this study (0.047 for CLR and 0.048 for CVR) were under the recommended levels (higher than 0.45) for human consumption. In comparison to Maniaci et al. (2021), the findings were often below than recommendation. According to Abbas et al. (2021), these cheeses had a PUFA/SFA value of 0.04, which is similar to what was found in our investigation. The MUFA/SFA ratio was higher for CVR (0.50) than for CLR (0.45), and these results are consistent with those found by Maniaci et al. (2021).

5. Conclusion

Based on the data obtained, the fresh cheese CVR made with cardoon flowers (*C. cardunculus*) differed significantly from the fresh cheese (CLR) made with lamb rennet for the most physicochemical parameters, including pH, acidity and dry matter, except for fat and protein contents. This study showed that the FA levels and lipid quality indices indicate practically the same values for all cheese samples (CVR and CLR). Finally, cheese (*Jben*) made with cardoon flowers had promising cheese characteristics when compared to CLR. This indicates that plant coagulants can be used and further valued for making a suitable *Jben* cheese suggesting that this milk-clotting ingredient can be a good substitute for animal rennet.

CRedit authors' contributions

Rachid Tabet (Conceptualization; Methodology; Data curation; Resources; Writing – original draft; Writing – reviewing & editing). Abdelbasset Mechai (Resources; Methodology; Writing – reviewing & editing). Zidane Branes (Methodology; Writing – reviewing & editing). Haroun Chenchoumi (Formal analysis; Visualization; Writing – reviewing & editing).

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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References

- Abbas, K.A., Abdelmontaleb, H.S., Hamdy, S.M., Ait-Kaddour, A., 2021. Physicochemical, functional, fatty acids profile, health lipid indices, microstructure and sensory characteristics of walnut-processed cheeses. *Foods* 10 (10), 2274. <https://doi.org/10.3390/foods10102274>.
- Aoac, 2005. *Official methods of analysis of the association of official agricultural chemists international*. J. Assoc. Off. Agric. Chem. 41, 12.
- Aquilanti, L., Babini, V., Santarelli, S., Osimani, A., Petruzzelli, A., Clementi, F., 2011. Bacterial dynamics in a raw cow's milk Caciotta cheese manufactured with aqueous extract of *Cynara cardunculus* dried flowers. *Lett. Appl. Microbiol.* 52 (6), 651–659. <https://doi.org/10.1111/j.1472-765X.2011.03053.x>.
- Barać, M., Kresojević, M., Špirović-Trifunović, B., Pešić, M., Vučić, T., Kostić, A., Despotović, S., 2018. Fatty acid profiles and mineral content of Serbian traditional white brined cheeses. *Mljekarstvo* 68 (1), 37–45. <https://hrcak.srce.hr/file/283669>.
- Benamara, R.N., Gemelas, L., Ibrri, K., Moussa-Boudjemaa, B., Demarigny, Y., 2016. Sensory, microbiological and physico-chemical characterization of *Klila*, a traditional cheese made in the south-west of Algeria. *Afr. J. Microbiol. Res.* 10 (41), 1728–1738. <https://doi.org/10.5897/ajmr2016.8264>.
- Benheddi, W., Hellal, A., 2019. Technological characterization and sensory evaluation of a traditional Algerian fresh cheese clotted with *Cynara cardunculus* L. flowers and lactic acid bacteria. *J. Food Sci. Technol.* 56 (7), 3431–3438. <https://doi.org/10.1007/s13197-019-03828-0>.
- Benyagoub, E.H., Guessas, B., Ayat, M., Sanebaoui, B., 2016. Physicochemical and bacteriological properties of some Algerian traditional dairy products 'Klila and Jben' marketed in southwest of Algeria and their impact on consumers' health. *Int. J. Adv. Res.* 4 (12), 760–768. <https://doi.org/10.21474/ijar01/2457>.
- Benyahia, F.A., Zitoun, O.A., Meghzili, B., Foufou, E., Zidoune, M.N., 2021. Use of *Pergularia tomentosa* plant enzymatic coagulant system in fresh cheese-making. *Food Nutr. Sci.* 12 (11), 1028–1040. <https://doi.org/10.4236/fns.2021.1211076>.
- Białek, A., Białek, M., Lepionka, T., Czerwonka, M., Czaderna, M., 2020. Chemometric analysis of fatty acids profile of ripening cheeses. *Molecules* 25 (8), 1814. <https://doi.org/10.3390/molecules25081814>.
- Buccioni, A., Mannelli, F., Daglio, M., Rapaccini, S., Scicutella, F., Minieri, S., 2022. Influence of milk quality and cheese-making procedure on functional fatty acid transfer in three Italian dairy products: mozzarella, Raveggiolo and Ricotta. *Lebensm. Wiss. Technol.* 163, 113476. <https://doi.org/10.1016/j.lwt.2022.113476>.
- Calder, P.C., 2012. Mechanisms of action of (n-3) fatty acids. *J. Nutr.* 142 (3), 592–599. <https://doi.org/10.3945/jn.111.155259>.
- Çetinkaya, A., Oz, F., 2018. Changes in cholesterol and free fatty acid content of Kars Gravyer Cheese (A Turkish dairy product produced by the traditional method). *Ukrainian food journal* 7 (3), 409–420.
- Chen, J., Liu, H., 2020. Nutritional indices for assessing fatty acids: a mini-review. *Int. J. Mol. Sci.* 21 (16), 5695. <https://doi.org/10.3390/ijms21165695>.
- Cruciata, M., Sannino, C., Ercolini, D., Scatassa, M.L., De Filippis, F., Mancuso, I., et al., 2014. Animal rennets as sources of dairy lactic acid bacteria. *Appl. Environ. Microbiol.* 80 (7), 2050–2061. <https://doi.org/10.1128/aem.03837-13>.
- Czyżak-Runowska, G., Wójtowski, J.A., Gogól, D., Wojtczak, J., Skrzypczak, E., Stanisławski, D., 2020. Properties of rennet cheese made from whole and skimmed summer and winter milk on a traditional polish dairy farm. *Animals* 10 (10), 1794. <https://doi.org/10.3390/ani10101794>.
- Dahou, A., Homrani, A., Bensaleh, F., Medjahed, M., 2015. La microflore lactique d'un fromage traditionnel Algérien «type J'ben»: connaissance des écosystèmes microbiens laitiers locaux et de leurs rôles dans la fabrication des fromages. *Afrique Sci.* 11 (6), 1–13.
- De la Haba Ruiz, M.A., Ruiz Pérez-Cacho, P., Dios Palomares, R., Galán-Soldevilla, H., 2016. Classification of artisanal Andalusian cheeses on physicochemical parameters applying multivariate statistical techniques. *Dairy Sci. Technol.* 96 (1), 95–106. <https://doi.org/10.1007/s13594-015-0242-5>.
- De Lorgeil, M., Renaud, S., Salen, P., Monjaud, I., Mabelle, N., Martin, J.L., et al., 1994. Mediterranean alpha-linolenic acid-rich diet in secondary prevention of coronary heart disease. *Lancet* 343 (8911), 1454–1459. [https://doi.org/10.1016/s0140-6736\(94\)92580-1](https://doi.org/10.1016/s0140-6736(94)92580-1).
- De Marchi, M., Bittante, G., Dal Zotto, R., Dalvit, C., Cassandro, M., 2008. Effect of Holstein Friesian and Brown Swiss breeds on quality of milk and cheese. *J. Dairy Sci.* 91 (10), 4092–4102. <https://doi.org/10.3168/jds.2007-0788>.
- Dopieralska, P., Barłowska, J., Teter, A., Król, J., Brodziak, A., Domaradzki, P., 2020. Changes in fatty acid and volatile compound profiles during storage of smoked cheese made from the milk of native polish cow breeds raised in the low beskids. *Animals* 10 (11), 2103. <https://doi.org/10.3390/ani10112103>.
- El-Galeel, A., Ali, A.A., El-Zawahry, A.A., 2017. Changes in physico chemical characteristics, proteolysis and organoleptic properties of edam cheese made with added aqueous extract of cardoon (*Cynara cardunculus*) and ginger (*Zingiber officinale*) during ripening. *Zagazig J. Agric. Res.* 44 (5), 1797–1808. <https://doi.org/>

- 10.21608/zjar.2017.52251.
- El-Kholly, A.M., 2015. Ras cheese making with vegetable coagulant – A comparison with calf rennet. *World J. Dairy Food Sci.* 10 (1), 82–89. <https://doi.org/10.5829/idosi.wjdfs.2015.10.1.95110>.
- Esmaeilzadeh, P., Ehsani, M.R., Mizani, M., Givianrad, M.H., 2021. Characterization of a traditional ripened cheese, Kurdish Kope: lipolysis, lactate metabolism, the release profile of volatile compounds, and correlations with sensory characteristics. *J. Food Sci.* 86 (8), 3303–3321. <https://doi.org/10.1111/1750-3841.15830>.
- Falchero, L., Lombardi, G., Gorlier, A., Lonati, M., Odoardi, M., Cavallero, A., 2010. Variation in fatty acid composition of milk and cheese from cows grazed on two alpine pastures. *Dairy Sci. Technol.* 90 (6), 657–672. <https://doi.org/10.1051/dst/2010035>.
- Fogeiro, É., Barracosa, P., Oliveira, J., Wessel, D.F., 2020. Influence of cardoon flower (*Cynara cardunculus* L.) and flock lactation stage in PDO Serra da Estrela cheese. *Foods* 9 (4), 386. <https://doi.org/10.3390/foods9040386>.
- Formaggioni, P., Summer, A., Malacarne, M., Franceschi, P., Mucchetti, G., 2015. Italian and Italian-style hard cooked cheeses: predictive formulas for Parmigiano-Reggiano 24-h cheese yield. *Int. Dairy J.* 51, 52–58. <https://doi.org/10.1016/j.idairyj.2015.07.008>.
- Fox, P.F., Guinee, T.P., Cogan, T.M., Mc Sweeney, P.L.H., 2017. *Fundamentals of Cheese Science*. Springer, New York. <https://doi.org/10.1007/978-1-4899-7681-9>.
- García, V., Rovira, S., Teruel, R., Boutoail, K., Rodríguez, J., Roa, L., López, M.B., 2012. Effect of vegetable coagulant, microbial coagulant and calf rennet on physicochemical, proteolysis, sensory and texture profiles of fresh goats cheese. *Dairy Sci. Technol.* 92 (6), 691–707. <https://doi.org/10.1007/s13594-012-0086-1>.
- Giosuè, C., D'Agostino, F., Maniaci, G., Avellone, G., Sciortino, M., De Caro, V., et al., 2022. Persistent organic pollutants and fatty acid profile in a typical cheese from extensive farms: First assessment of human exposure by dietary intake. *Animals* 12 (24), 3476. <https://doi.org/10.3390/ani12243476>.
- Gomes, S., Belo, A.T., Alvarenga, N., Dias, J., Lage, P., Pinheiro, C., et al., 2019. Characterization of *Cynara cardunculus* L. flower from Alentejo as a coagulant agent for cheesemaking. *Int. Dairy J.* 91, 178–184. <https://doi.org/10.1016/j.idairyj.2018.09.010>.
- Gómez-Cortés, P., Juárez, M., de la Fuente, M.A., 2018. Milk fatty acids and potential health benefits: An updated vision. *Trends Food Sci. Technol.* 81, 1–9. <https://doi.org/10.1016/j.tifs.2018.08.014>.
- Gulzar, N., Rafiq, S., Nadeem, M., Imran, M., Khaliq, A., Muqada Sleem, I., Saleem, T., 2019. Influence of milling pH and storage on quality characteristics, mineral and fatty acid profile of buffalo Mozzarella cheese. *Lipids Health Dis.* 18 (1), 1–8. <https://doi.org/10.1186/s12944-019-0976-9>.
- Gutiérrez-Peña, R., Avilés, C., Galán-Soldevilla, H., Polvillo, O., Ruiz Pérez-Cacho, P., Guzmán, J.L., et al., 2021. Physicochemical composition, antioxidant status, fatty acid profile, and volatile compounds of milk and fresh and ripened ewes' cheese from a sustainable part-time grazing system. *Foods* 10 (1), 80. <https://doi.org/10.3390/foods10010080>.
- Hanuš, O., Samková, E., Křížová, L., Hasoňová, L., Kala, R., 2018. Role of fatty acids in milk fat and the influence of selected factors on their variability—a review. *Molecules* 23 (7), 1636. <https://doi.org/10.3390/molecules23071636>.
- Hashim, M.M., Dong, M., Iqbal, M.F., Li, W., Chen, X., 2011. Ginger protease used as coagulant enhances the proteolysis and sensory quality of Peshawari cheese compared to calf rennet. *Dairy Sci. Technol.* 91 (4), 431–440. <https://doi.org/10.1007/s13594-011-0021-x>.
- Hayaloglu, A.A., 2017. Cheese varieties ripened under brine. In: McSweeney, P.L.H., Fox, P.F., Cotter, P.D., Everett, D.W. (Eds.), *Cheese*. fourth ed., Academic Press, San Diego, p. 997–1040. <https://doi.org/10.1016/B978-0-12-417012-4.00039-9>chap. 39.
- Hayaloglu, A.A., Guven, M., Fox, P.F., 2002. Microbiological, biochemical and technological properties of Turkish White cheese 'Beyaz Peynir'. *Int. Dairy J.* 12 (8), 635–648. [https://doi.org/10.1016/S0958-6946\(02\)00055-9](https://doi.org/10.1016/S0958-6946(02)00055-9).
- Hirigoyen, D., De Los Santos, R., Calvo, M.F., González-Revello, A., Constantin, M., 2018. Chemical composition and seasonal changes in the fatty acid profile of Uruguayan “Colonia” Cheeses. *Grasas Aceites* 69 (2), e254–e254. <https://doi.org/10.3989/gya.1217172>.
- Holub, D.J., Holub, B.J., 2004. Omega-3 fatty acids from fish oils and cardiovascular disease. *Mol. Cell. Biochem.* 263 (1), 217–225. <https://doi.org/10.1023/b:mcbi.0000041863.11248.8d>.
- Ioannidou, M.D., Maggira, M., Samouris, G., 2022. Physicochemical characteristics, fatty acids profile and lipid oxidation during ripening of graviera cheese produced with raw and pasteurized milk. *Foods* 11 (14), 2138. <https://doi.org/10.3390/foods11142138>.
- Irlinger, F., Mounier, J., 2009. Microbial interactions in cheese: implications for cheese quality and safety. *Curr. Opin. Biotechnol.* 20 (2), 142–148. <https://doi.org/10.1016/j.copbio.2009.02.016>.
- Islam, M.A., Basunia, M.H.K., Rahman, A., Bari, M.S., Rahman, M.F., Mannan, M.A., Datta, T.K., 2021. Effect of coagulants on the chemical and microbial quality of fresh cheese. *Bangladesh J. Anim. Sci.* 50 (2), 73–79. <https://doi.org/10.3329/bjas.v50i2.58024>.
- ISO, 2001. *Milk and Milk Products – Extraction Methods for Lipids and Liposoluble Compounds*. International Organization for Standardization (ISO), Geneva, Switzerland. ISO 14156).
- ISO, 2002a. *Milk Fat – Preparation of Fatty Acid Methyl Esters*. International Organization for Standardization (ISO), Geneva, Switzerland. ISO 15884).
- ISO, 2002b. *IDF. Milk Fat-Determination of the FA Composition by Gas-Chromatography*, vol. 184. International Organization for Standardization (ISO), Geneva, Switzerland. (ISO 15885), 2002.
- ISO, 2004. *Cheese and Processed Cheese – Determination of the Total Solids Content (Reference Method)*, International Organization for Standardization (ISO), Geneva, Switzerland, p. 15. (ISO 5534).
- ISO, 2014. *Milk and Milk Products – Determination of Nitrogen Content – Part 1: Kjeldahl Principle and Crude Protein Calculation*, International Organization for Standardization (ISO), Geneva, Switzerland, p. 30. ISO 8968-1).
- Ivanova, A., Hadzhinikolova, L., 2015. Evaluation of nutritional quality of common carp (*Cyprinus carpio* L.) lipids through fatty acid ratios and lipid indices. *Bulg. J. Agric. Sci* 21, 180–185.
- Jeanet, R., Croguennec, T., Schuch, P., Brulé, G., 2007. *Science des aliments: Biochimie – Microbiologie – Procédés – Produits*, vol. 2. Technologie des produits alimentaires, Tech et Doc, Paris, p. 456p.
- Kajak-Siemaszko, K., Zielińska, D., Lepecka, A., Jaworska, D., Okoń, A., Neffe-Skocińska, K., et al., 2022. Effect of Lactic Acid Bacteria on Nutritional and Sensory Quality of Goat Organic Acid-Rennet Cheeses. *Appl. Sci.* 12, 8855. <https://doi.org/10.3390/app12178855>.
- Kanekanian, A., 2014. The health benefits of bioactive compounds from milk and dairy products. In: Kanekanian, A. (Ed.), *Milk and Dairy Products as Functional Foods*. Wiley, p. 1–22. <https://doi.org/10.1002/9781118635056.ch1>.
- Kilcawley, K.N., Mannion, D.T., 2017. Free fatty acids quantification in dairy products. In: Catala, A. (Ed.), *Fatty Acids*. IntechOpen, London, UK, p. 209–220. <https://doi.org/10.5772/intechopen.69596>.
- Koirala, A., 2021. Comparative study on physico-chemical and microbiological properties of soft cheese prepared by using crude Aank (*Calotropis gigantea*) and jackfruit (*Artocarpus heterophyllus*) proteases. In: *Doctoral Dissertation*. Tribhuvan University, Dharan.
- Lahsoui, S., 2009. *Étude du procédé de fabrication d'un produit laitier traditionnel Algérien (Kilila)*. Doctoral dissertation, Univ. Batna, Algeria.
- Leksir, C., Boudalia, S., Moujahed, N., Chemmam, M., 2019. Traditional dairy products in Algeria: case of Klila cheese. *J. Ethnic. Food.* 6 (1), 7. <https://doi.org/10.1186/s42779-019-0008-4>.
- Liu, X., Wu, Y., Guan, R., Jia, G., Ma, Y., Zhang, Y., 2021. Advances in research on calf rennet substitutes and their effects on cheese quality. *Food Res. Int.* 149, 110704. <https://doi.org/10.1016/j.foodres.2021.110704>.
- Manca, G., Ru, A., Siddi, G., Mocchi, A.M., Murittu, G., De Santis, P.L., 2020. Biogenic amines content in Fiore Sardo cheese in relation to free amino acids and physicochemical characteristics. *Italy J. Food Safety.* 9 (1), 8457. <https://doi.org/10.4081/jfs.2020.8457>.
- Maniaci, G., Di Grigoli, A., Bonanno, A., Giosuè, C., Iardi, V., Alabiso, M., 2021. Fatty acids as biomarkers of the production season of Caciocavallo Palermitano cheese. *Animals* 11 (9), 2675. <https://doi.org/10.3390/ani11092675>.
- Markiewicz-Kęszczyca, M., Czyżak-Runowska, G., Lipińska, P., Wójt, J., 2013. Fatty acid profile of milk a review. *Bull. Vet. Inst. Pulawy* 57, 135–139. <https://doi.org/10.2478/bvip-2013-0026>.
- Martino, C., Ianni, A., Grotta, L., Pomilio, F., Martino, G., 2019. Influence of zinc feeding on nutritional quality, oxidative stability and volatile profile of fresh and ripened ewes' milk cheese. *Foods* 8 (12), 656. <https://doi.org/10.3390/foods8120656>.
- Maskey, B., Shrestha, N.K., 2020. Optimization of crude papaya (*Carica papaya*) prestase in soft-unripened cheese preparation. *J. Food Sci. Technol. Nepal* 12 (12), 1–8. <https://doi.org/10.3126/jfstn.v12i12.30139>.
- Medjoudj, H., Aouar, L., Derouiche, M., Choiset, Y., Haertlé, T., Chobert, J.M., et al., 2020. Physicochemical, microbiological characterization and proteolysis of

- Algerian traditional *Bouhezza* cheese prepared from goat's raw milk. *Anal. Lett.* 53 (6), 905–921. <https://doi.org/10.1080/00032719.2019.1685531>.
- Michikawa, M., 2003. The role of cholesterol in pathogenesis of Alzheimer's disease. *Mol. Neurobiol.* 27 (1), 1–12. <https://doi.org/10.1385/mn:27:1:1>.
- Miličević, D., Krešić, G., Vranić, D., Lešić, T., Nedeljković-Trailović, J., Janković, S., Pleadin, J., 2022. Characterization and nutritional assessment of traditional dairy products from the Zlatibor region, Republic of Serbia. *Veterinarska Stanica* 53 (1), 17–32. <https://doi.org/10.46419/vs.53.1.11>.
- Mladenović, K.G., Grujović, M.Ž., Kocić-Tanackov, S.D., Bulut, S., Iličić, M., Degenek, J., Semedo-Lemsaddek, T., 2022. Serbian traditional goat cheese: physico-chemical, sensory, hygienic and safety characteristics. *Microorganisms* 10 (1), 90. <https://doi.org/10.3390/microorganisms10010090>.
- Mutwedu, V.B., Ayagirwe Rodrigue Basengere, Y., Mugumaarhahama, G.B., Barume, A., Matendo, R., Bisimwa, E., et al., 2018. Effets des techniques de transformation sur la qualité du fromage blanc traditionnel «Mashanza» produit au Sud-Kivu, RD Congo. *J. Animal Plant Sci.* 38 (1), 6097–6111.
- Nair, M.K.M., Joy, J., Vasudevan, P., Hinckley, L., Hoagland, T.A., Venkitanarayanan, K.S., 2005. Antibacterial effect of caprylic acid and monocaprylin on major bacterial mastitis pathogens. *J. Dairy Sci.* 88 (10), 3488–3495. [https://doi.org/10.3168/jds.s0022-0302\(05\)73033-2](https://doi.org/10.3168/jds.s0022-0302(05)73033-2).
- Nielsen, S.S., 2017. Standard solutions and titratable acidity. In: Suzanne Nielsen, S. (Ed.), *Food Analysis Laboratory Manual*. Springer, Cham, p. 179–184. https://doi.org/10.1007/978-3-319-44127-6_21.
- Omri, B., Chalghoumi, R., Izzo, L., Ritieni, A., Lucarini, M., Durazzo, A., et al., 2019. Effect of dietary incorporation of linseed alone or together with tomato-red pepper mix on laying hens' egg yolk fatty acids profile and health lipid indexes. *Nutrients* 11 (4), 813. <https://doi.org/10.3390/nu11040813>.
- Osmari, E.K., Cecato, U., Macedo, F.A.F., Souza, N.E., 2011. Nutritional quality indices of milk fat from goats on diets supplemented with different roughages. *Small Rumin. Res.* 98 (1–3), 128–132. <https://doi.org/10.1016/j.smallrumres.2011.03.030>.
- Papademas, P., Bintsis, T. (Eds.), 2017. *Global Cheesemaking Technology: Cheese Quality and Characteristics*. Wiley, Chichester, UK. <https://doi.org/10.1002/9781119046165>.
- Paszczyk, B., Luczyńska, J., 2020. The comparison of fatty acid composition and lipid quality indices in hard cow, sheep, and goat cheeses. *Foods* 9 (11), 1667. <https://doi.org/10.3390/foods9111667>.
- Paszczyk, B., Polak-Śliwińska, M., Luczyńska, J., 2020. Fatty acids profile, trans isomers, and lipid quality indices in smoked and unsmoked cheeses and cheese-like products. *Int. J. Environ. Res. Publ. Health* 17 (1), 71. <https://doi.org/10.3390/ijerph17010071>.
- Paszczyk, B., Polak-Śliwińska, M., Zielak-Steciwo, A.E., 2022. Chemical composition, fatty acid profile, and lipid quality indices in commercial ripening of cow cheeses from different seasons. *Animals* 12 (2), 198. <https://doi.org/10.3390/ani12020198>.
- R Core Team, 2022. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. URL: <https://www.R-project.org>.
- Roseiro, L.B., Barbosa, M., Ames, J.M., Wilbey, R.A., 2003. Cheesemaking with vegetable coagulants—the use of *Cynara L.* for the production of ovine milk cheeses. *Int. J. Dairy Technol.* 56 (2), 76–85. <https://doi.org/10.1046/j.1471-0307.2003.00080.x>.
- Russo, G.L., 2009. Dietary n–6 and n–3 polyunsaturated fatty acids: from biochemistry to clinical implications in cardiovascular prevention. *Biochem. Pharmacol.* 77 (6), 937–946. <https://doi.org/10.1016/j.bcp.2008.10.020>.
- Santillo, A., Caroprese, M., Marino, R., Muscio, A., Sevi, A., Albenzio, M., 2007. Influence of lamb rennet paste on composition and proteolysis during ripening of Pecorino foggiano cheese. *Int. Dairy J.* 17 (5), 535–546. <https://doi.org/10.1016/j.idairyj.2006.06.024>.
- Senoussi, A., Rapisarda, T., Schadt, I., Chenchoumi, H., Saoudi, Z., Senoussi, S., et al., 2022. Formation and dynamics of aroma compounds during manufacturing-ripening of *Bouhezza* goat cheese. *Int. Dairy J.* 129 (1), 105349. <https://doi.org/10.1016/j.idairyj.2022.105349>.
- Senoussi, A., Schadt, I., Hioum, S., Chenchoumi, H., Saoudi, Z., Aissaoui Zitoun Hamama, O., Zidoune, M.N., Carpino, S., Rapisarda, T., 2021. Botanical composition and aroma compounds of semi-arid pastures in Algeria. *Grass Forage Sci.* 76 (2), 282–299. <https://doi.org/10.1111/gfs.12510>.
- Silva, M.D.F.S., Bessa, R., 2002. Effect of genotype, feeding system and slaughter weight on the quality of light lambs. *Livest. Prod. Sci.* 77, 187–194. [https://doi.org/10.1016/S0301-6226\(02\)00059-3](https://doi.org/10.1016/S0301-6226(02)00059-3).
- Simopoulos, A.P., 2002. The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomed. Pharmacother.* 56 (8), 365–379. [https://doi.org/10.1016/S0753-3322\(02\)00253-6](https://doi.org/10.1016/S0753-3322(02)00253-6).
- Simopoulos, A.P., 2006. Evolutionary aspects of diet, the omega-6/omega-3 ratio and genetic variation: nutritional implications for chronic diseases. *Biomed. Pharmacother.* 60 (9), 502–507. <https://doi.org/10.1016/j.biopha.2006.07.080>.
- Simopoulos, A.P., 2008. The importance of the omega-6/omega-3 fatty acid ratio in cardiovascular disease and other chronic diseases. *Exp. Biol. Med.* 233 (6), 674–688. <https://doi.org/10.3181/0711-mr-311>.
- Summer, A., Formaggioli, P., Franceschi, P., Di Frangia, F., Righi, F., Malacarne, M., 2017. Cheese as functional food: the example of parmigiano reggiano and grana padano. *Food Technol. Biotechnol.* 55 (3), 277–289. <https://doi.org/10.17113/ftb.55.03.17.5233>.
- Tadjine, D., Boudalia, S., Bousbia, A., Gueroui, Y., Symeon, G., Mebirouk Boudechiche, L., et al., 2020. Milk heat treatment affects microbial characteristics of cows' and goats' "Jben" traditional fresh cheeses. *Food Sci. Technol.* 41, 136–143. <https://doi.org/10.1590/fst.00620>.
- Tofalo, R., Schirone, M., Fasoli, G., Perpetuini, G., Patrignani, F., Manetta, A.C., et al., 2015. Influence of pig rennet on proteolysis, organic acids content and microbiota of Pecorino di Farindola, a traditional Italian Ewe's raw milk cheese. *Food Chem.* 175, 121–127. <https://doi.org/10.1016/j.foodchem.2014.11.088>.
- Ulbricht, T.L.V., Southgate, D.A.T., 1991. Coronary heart disease: seven dietary factors. *Lancet* 338 (8773), 985–992. [https://doi.org/10.1016/0140-6736\(91\)91846-M](https://doi.org/10.1016/0140-6736(91)91846-M).
- Uzun, P., Serrapica, F., Masucci, F., Assunta, B.C.M., Yildiz, H., Grasso, F., Di Francia, A., 2020. Diversity of traditional Caciocavallo cheeses produced in Italy. *Int. J. Dairy Technol.* 73 (1), 234–243. <https://doi.org/10.1111/1471-0307.12640>.
- Wei, T., Simko, V., 2021. R Package 'corrplot': Visualization of a Correlation Matrix. Available from: Version 0.92. <https://github.com/taiyun/corrplot>.
- Willett, W.C., 2007. The role of dietary n-6 fatty acids in the prevention of cardiovascular disease. *J. Cardiovasc. Med.* 8, S42–S45. <https://doi.org/10.2459/01.jcm.0000289275.72556.13>.
- Willett, W.C., Stampfer, M.J., Manson, J.E., Colditz, G.A., Speizer, F.E., Rosner, B.A., et al., 1993. Intake of trans fatty acids and risk of coronary heart disease among women. *Lancet* 341 (8845), 581–585. [https://doi.org/10.1016/0140-6736\(93\)90350-p](https://doi.org/10.1016/0140-6736(93)90350-p).
- Wood, J.D., Richardson, R.I., Nute, G.R., Fisher, A.V., Campo, M.M., Kasapidou, E., et al., 2004. Effects of fatty acids on meat quality: a review. *Meat Sci.* 66 (1), 21–32. [https://doi.org/10.1016/S0309-1740\(03\)00022-6](https://doi.org/10.1016/S0309-1740(03)00022-6).