European Food Research and Technology (2021) 247:2211–2225
https://doi.org/10.1007/s00217-021-03780-4

ORIGINAL PAPER

Effect of adjunct starter culture on the quality of reduced fat, white, brined goat cheese: part I. Assessment of chemical composition, proteolysis, lipolysis, texture and sensory attributes

A. Zaravela[1] · S. Kontakos[2] · A. V. Badeka[1] · M. G. Kontominas[1]

Received: 9 April 2021 / Revised: 11 May 2021 / Accepted: 15 May 2021 / Published online: 24 May 2021

Abstract
Full fat (4.3%) and two reduced fat (2.5% and 1.4%) milk were used to prepare white brined goat cheeses with the addition of (i) mixed mesophilic/thermophilic (MT1) or (ii) mesophilic/thermophilic (MT1) plus thermophilic adjunct culture (LH-B02) as starters. Chemical composition, proteolysis and lipolysis were determined, followed by texture profile analysis and sensory evaluation of cheese samples. The combination of main and adjunct culture resulted in higher values of proteolysis parameters (water-soluble nitrogen—% WSN, trichloroacetic acid-soluble nitrogen—% TCA-SN and phosphotungstic acid-soluble nitrogen—% PTA-SN) and lower values of all textural properties. The main free fatty acids of all mature cheese samples were hexanoic (short chain acid), lauric and myristic acid (medium chain acids) and palmitic acid (long chain acid). Reduced fat, white, brined cheeses made with 1.4 and 2.5% milk fat and both main and adjunct cultures were judged as acceptable by panelists for a 12-month period without any off-flavor development.

Keywords Reduced fat · White brined cheese · Adjunct culture · Chemical composition · Proteolysis and lipolysis · Texture profile analysis · Sensory evaluation

Introduction

A number of studies on the nutritional value of goat milk and its products have been published recently [1–5]. Several studies have revealed that goat milk is a nutritional and therapeutic food being an excellent raw material for development of functional foods [1, 5]. Kalyan et al. [3] reported that goat milk fat and casein possess anti-hypercholesterolaemic and antioxidative properties. Goat milk caseins, also, tend to be more efficiently digested compared to cow milk by infants and young children [1, 2].

Goat farming has been one of the earliest agricultural activities in Greece. Currently, Greece has the largest goat population in the EU. According to FAOSTAT [6] Greece held third place in goat milk production in 2018 (338,400 tons) with France holding first place (648,370 tons) and Spain holding second place (515,550 tons) in the European Union.

Brined cheeses are cheeses that are ripened and preserved in brine. Traditionally brined cheeses are produced in numerous Balkan and Mediterranean countries [7]. White, brined goat cheese is prepared in various parts of Greece and ripens over 2 months before it can be consumed [8]. Its commercial shelf life under refrigeration is ca. 18 months. White, brined goat cheese is similar to Feta cheese differing in texture, flavor and chemical composition. That is, Feta cheese, a PDO product of Greece since 2002, is made of ewe milk or mixtures with goat milk, with the latter not exceeding 30% of the total milk used.

Fat is a functional constituent that plays an essential role in flavor, texture and color development in cheeses [9]. Nowadays, consumers are looking for healthy reduced and low-fat products. Unfortunately, reduced and low-fat cheeses are often less sensorially acceptable due to the lack of flavor and texture defects compared to the full-fat cheeses [10]. In order to overcome such defects, three basic strategies have been

✉ A. V. Badeka
abadeka@uoi.gr

✉ M. G. Kontominas
mkontomi@uoi.gr

[1] Laboratory of Food Chemistry, Department of Chemistry, University of Ioannna, 45110 Ioannina, Greece

[2] Department of Social Administration and Political Science, Democritus University of Thrace, 69100 Komotini, Greece

suggested: (i) fat replacers, (ii) changes in cheese production processes and (iii) adjunct cultures. Fat replacers and other additives often cause off-flavors in the final product [9, 11]. On the other hand, various adjunct cultures are capable of producing enzymes with specificity to hydrolyze caseins, leading to the release of bioactive peptides and the formation of additional volatile compounds [12].

Cheese ripening is a complex procedure that affects flavor and texture of all types of cheeses. The main biochemical reactions in the development of flavor in cheese during ripening are proteolysis, lipolysis and glycolysis [13, 14]. Protein degradation affects cheese flavor and texture. Proteolytic agents in cheese originate from (i) the coagulant (rennet), (ii) indigenous and endogenous proteinases from the milk (plasmin and heat-stable proteinases from psychrotrops), (iii) the starter culture and (iv) the adjunct starter and non-starter lactic acid bacteria (proteolytic enzymes). For the development of an acceptable and characteristic flavor according to type of cheese, a well-balanced breakdown of casein into small peptides and amino acids is necessary, since these products are flavor compounds or act as precursors for the formation of flavor compounds during the ripening process. Although large peptides do not contribute directly to cheese flavor, they are very important for the development of the desirable texture [7, 12, 15].

Lipolysis in cheese is caused by lipolytic enzymes originating from the following sources: (i) milk indigenous lipases (ii) rennet pregastric esterase, (iii) starter lactic acid bacteria (LAB), (iv) starter cultures and non-starter lactic acid bacteria (NSLAB) via lipolytic and/ or esterolytic enzymes they contain and (v) exogenous lipase preparations [16]. Short-chain fatty acids produced during lipolysis, contribute to the characteristic flavor of cheese. Cheese flavor is also affected by acetic acid, propanoic acid and some FFAs produced from lactose fermentation and amino acid degradation by LAB and NSLAB while most free fatty acids $C_{4:0}$–$C_{20:0}$ arise from hydrolysis of milk triglycerides during aging [17]. The free fatty acids comprise precursors for a variety of flavor compounds such as methylketones, secondary alcohols, esters and lactones [18].

Currently, there is a wide interest in reduced and low-fat cheeses [19–23] especially in low-fat feta type cheese [24, 25] such as reduced fat, brined goat cheese. In order to prepare low-fat cheeses, the fat content of milk used should range from < 0.5% to approximately 1.8%, w /v [11].

In the present study, a series of fat-reduced, white, brined goat cheeses were prepared using 90% goat milk and 10% ewe milk. In order to improve cheese flavor, the addition of a mesophilic/thermophilic adjunct culture was investigated. To the best of our knowledge there are no reports in the literature on the improvement of reduced-fat white, brined goat cheese flavor using adjunct starter cultures. Furthermore, for the first time, proteolysis, lipolysis, texture and sensory profile of reduced-fat, white, brined goat cheese is investigated over a 12-month period, both the above comprising the novelty of the study.

Materials and methods

Milk

Reduced fat, white, brined cheese samples were prepared using a mixture of goat and ewe milk at a rate of 90% and 10%, respectively. Milk originated from the region of Epirus, Greece and was donated by Dodoni dairy S.A., Ioannina, Greece. Table 1 shows the physicochemical characteristics of milk used for cheese production.

Cultures

The CHOOZIT MT1 freeze-dried culture is a commercially available mesophilic/thermophilic culture containing *Lactococcus lactis* subsp. *lactis, Lactococcus lactis* subsp. *cremoris, Streptococcus salivarius* subsp. *Thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* (Danisco, France). MT1 was used as the main culture. The adjunct culture was the freeze-dried, LH-B02 culture containing *Lactobacillus helveticus* (Chr. Hansen, Denmark).

Cheese making and sampling

The cheese samples were prepared in the Dairy School of Ioannina using 150 kg milk to formulate each of the six batches (two batches for each fat content). Cheese making was carried out according to the official quality control scheme used for commercial production of feta cheese by Dodoni Dairy Co. The milk was pre-heated at 40 °C and standardized by centrifugation in order to prepare full-fat and reduced-fat cheeses. Batches of milk were standardized

Table 1 Composition and physicochemical properties of milk used for cheese production

Batch	RF2	RF1	FF
Fat (%, w/v)	1.40 ± 0.00^{a}	2.50 ± 0.08^{b}	4.30 ± 0.07^{c}
Protein (%, w/v)	4.21 ± 0.03^{a}	4.16 ± 0.04^{a}	4.12 ± 0.05^{a}
Lactose (%, w/v)	4.83 ± 0.04^{b}	4.69 ± 0.05^{a}	4.62 ± 0.03^{a}
Acidity (°D)	15.50 ± 0.15^{b}	15.00 ± 0.09^{a}	15.00 ± 0.20^{a}
pH	6.69 ± 0.04^{a}	6.68 ± 0.05^{a}	6.65 ± 0.07^{a}

Data are expressed as means ± standard deviation. All samples were analyzed in duplicate for each cheese making session. Means with different superscript letter in the same column of each factor were significantly different ($P < 0.05$)

FF full-fat control (4.3%, w /v), *RF1* reduced fat (2.5%, w /v), *RF2* reduced fat (1.4%, w /v)

to 4.3%, w /v (FF, control), 2.5%, w /v (RF1) and 1.4%, w /v (RF2) fat (Table 1). Each batch of milk was pasteurized in a double-walled stainless-steel vat type pasteurizer at 63 °C for 30 min and then cooled to 34 – 35 °C. A quantity of 1.03 g of Normal starter culture MT1 (Danisco France) was added to each batch. The adjunct culture (0.13 g) LH-B02 (Chr. Hansen, Denmark) was added only to three of the six batches (one for each FF, RF1 and RF2). The starters were allowed to ripen for 20 min. Then $CaCl_2$ solution 50% (20 mL/100 kg) was added to the mixture. Calf rennet (Naturen Extra 220, Chr. Hansen, Denmark) was added to achieve curdling in approximately 45 min at 34–35 °C. After coagulation, the curd was cut into cubes of dimensions 3 × 3 × 3 cm and left to rest for 15 min. The sliced curd was transferred into perforated inox parallelepiped molds for draining. The molds were inverted twice: the first time after 2 h and the second after an additional hour. After the last inversion, the molds were left for about 12 h at 16 °C to complete draining. The curd was removed from each mold, cut and transferred into individual parallelepiped plastic vessels of dimensions 35 × 25 × 25 cm. After 24 h, granular salt was added in an amount of 25 g/kg of cheese weight. On the next day, the surface of the cheese samples was cleaned manually from excess salt and cheese was transferred into parallelepiped metal cans of dimensions 17 × 23.5 × 23.5 cm. Brine solution was added to the cans at a concentration of 65 g/kg. The cans were then transferred to the ripening room (at a temperature of 16–18 °C). The cheeses remained there until the pH of cheese dropped to a value of 4.6 and only for full-fat samples the moisture content to 56 g/100 g. This was achieved in approximately 9–11 days. Then the cans were sealed and transferred into the refrigerator at 4 ± 2 °C and kept there for up to a year. The experiment was replicated twice on different occasions under identical conditions. All microbiological analyses of raw material and produced cheese were run by DODONI Dairy Co. following their official quality control scheme for commercial feta cheese production.

Physico-chemical analysis

pH of milk and cheeses was determined using a model WTW (SenTix SP-DIN, 2010 pH meter (Wissenschaftlich–Technische Werkstätten GmbH, Germany). The titratable acidity of milk was determined by the Dornic method [26].

All cheese samples (full and reduced fat) were analyzed for fat, protein, moisture, salt content, total nitrogen (TN), water-soluble nitrogen (% WSN), nitrogen-soluble in 12% trichloroacetic acid (% TCA-SN) and nitrogen-soluble in 5% phosphotungistic acid (% PTA-SN). Fat in milk and cheeses was determined by Gerber van Gulik method [26] and [27], respectively. Moisture in cheeses was determined according to the oven drying method [28]. Cheese salt content was determined by the Mohr method [29]. Milk protein content, total nitrogen (TN) and soluble nitrogen were determined according to the Kjeldahl method [30] using the Inkjel 1210 M apparatus (Behr Labor—Technik Gmb., Düsseldorf, Germany). WSN, TCA–SN and PTA–SN were determined as described by Michaelidou et al. [25]. Salt in moisture content was calculated according to Pappa et al. [31].

All determinations were run in duplicate per replicate ($n = 2 \times 2 = 4$).

Free fatty acids analysis

Lipolysis was assessed after extraction of cheese lipids, isolation of FFA and determination of their concentration by GC according to De Jong and Bandings [32]. A gas chromatograph Agilent 6820 GC (G1176A) model equipped with a FID was used with a capillary column length 30 m, inner diameter 0.25 mm, coated with FFA phase OV-351 film 0.25 μm nitroterephthalic acid modified polyethylene glycol (122—3232 Agilent J & W GC Columns USA). Gas chromatography oven conditions were programmed from 65 to 240 °C at a rate of 10 °C min^{-1} and then held at 240 °C for 20 min. The FID temperature was 250 °C and helium was carrier gas and nitrogen was auxiliary gas at flow rate 1 ml min^{-1} and 30 ml min^{-1}, respectively, and the split ratio at 1:10. The identification of FFA of the cheese samples was performed via comparison of the retention times of the unknown FFA to those of known standards (Merck Schuchardt OHG, Germany) under identical conditions. The quantification of the FFA of all cheese samples was determined using $C_{9:0}$ as internal standard [32] and processing the chromatograms with the software (Agilent Cerity A.04.07, USA).

All determinations were run in duplicate per replicate ($n = 2 \times 2 = 4$).

Texture evaluation

Texture profile analysis (TPA) of cheeses was performed using an Instron Universal Testing Instrument model 4411, (High Wycombe, UK) with a cylindrical plunger 35 mm in diameter. The cheese samples were cut into cube-shaped pieces (with a cube edge of 20 mm) using a stainless steel cutter. Sampling was carried out on several parts of the cheese body, in order to prepare a representative sample. All measurements were made at room temperature (ca. 20 °C). The compression of the sample was set at 70% in one cycle (bite) and the force/time curve was constructed. The operating conditions were a) compressive load cell of 5 kN and b) crosshead speed of 30 mm/min. Hardness was defined as the peak force during the first compression cycle "first bite". Fracturability or brittleness was defined as the force at the first significant break in the curve [33].

Ten texture measurements were run per sample per replicate ($n = 10 \times 2 = 20$).

Sensory evaluation

The cheeses were evaluated by seven trained panelists on days 60, 120, 180, 270 and 365 of storage in individual booths according to the IDF method [34] as modified by Katsiari and Voutsinas [35]. The samples were cut into cube-shaped pieces and placed in 3-digit randomly coded plastic cups at room temperature for trial. The panelists were asked to eat a cracker biscuit and drink water between evaluation of samples. They evaluated flavor (odor and taste), texture (body) and appearance (exterior–interior). The scoring scale ranged between 0 and 100 with 60 being the lower acceptability limit. The individual scores of flavor and texture were multiplied by four and five, respectively. The total score was calculated by adding the individual scores of above attributes [35].

Statistical analysis

Data were subjected to analysis of variance (ANOVA) in order to determine the significance of the individual differences. Significant means were compared using the Tukey test at the level of $P = 0.05$. Pearson's correlation was performed among texture profile parameters, chemical composition, pH, proteolysis and sensory evaluation of the cheese samples after 120 days at the level of $P = 0.05$ and $P = 0.01$. All data analysis was performed using the SPSS statistical package (version 20, IBM Corp., New York, USA).

Results and discussion

Chemical composition

Results on chemical composition of the white, brined cheese samples are given in Table 2. According to the Greek Foods and Beverages Codex [8], good quality white, brined, full-fat cheeses, should contain at least 43% (w/w) of fat in dry matter (FDM) and maximum moisture of 56% (w/w). In the present study the FDM was 53.21% (23.68% as is basis) while moisture was 55.50% for the full-fat (FF) cheeses samples. Respective values of the reduced fat samples (RF1) were 36.15% (13.75% as is basis) and 62.01% and for reduced-fat RF2 were 25.92% (9.13% as is basis) and 64.80%. These values correspond to a reduction of fat of ca. 41.9% for the RF1 and 61.4% for the RF2 cheeses samples.

Table 2 Effect of milk fat content, starter culture and ripening time on the chemical composition of white brined goat cheeses, analyzed by ANOVA

Factors		Fat (%, w/w)	FDM (%, w/w)	MNFS (%, w/w)	Moisture (%, w/w)	Protein (%, w/w)	Salt (%, w/w)	S/M (%, w/v)	pH
Treatment	FF	23.68^{a}	53.21^{a}	72.72^{a}	55.50^{a}	16.62^{a}	2.59^{a}	4.71^{a}	4.42^{a}
($n = 64$)	RF1	13.75^{b}	36.15^{b}	71.89^{b}	62.01^{b}	17.50^{b}	2.96^{b}	4.80^{a}	4.45^{a}
	RF2	9.13^{c}	25.92^{c}	71.30^{c}	64.80^{c}	19.72^{c}	3.08^{c}	4.78^{a}	4.51^{b}
MSE		0.190	0.690	0.169	0.211	0.134	0.010	0.026	0.005
MSD		4.62	10.23	0.585	2.790	0.877	0.121	0.021	0.030
Culture	M	15.57^{a}	38.50^{a}	71.97^{a}	60.80^{a}	18.18^{a}	2.86^{a}	4.74^{a}	4.46^{a}
($n = 96$)	MA	15.47^{a}	38.36^{a}	71.96^{a}	60.74^{a}	17.71^{b}	2.89^{a}	4.791^{a}	4.47^{a}
MSE		0.190	0.690	0.169	0.211	0.134	0.010	0.026	0.005
MSD		0.101	0.134	0.013	0.058	0.467	0.033	0.048	0.011
Days	1	14.12^{a}	37.75^{a}	74.18^{a}	63.66^{a}	18.87^{a}	–	–	4.87^{a}
($n = 24$)	9–11	15.41^{b}	38.30^{abc}	71.99^{b}	60.87^{b}	18.57^{ab}	2.73^{a}	4.48^{a}	4.42^{b}
	30	15.41^{b}	38.01^{ab}	71.64^{bc}	60.58^{bc}	18.12^{bc}	2.88^{b}	4.74^{b}	4.37^{b}
	60	15.63^{bc}	38.38^{abc}	71.72^{bc}	60.48^{bc}	17.84^{cd}	2.88^{b}	4.76^{b}	4.36^{b}
	120	15.54^{bc}	38.17^{ab}	71.59^{bc}	60.44^{bc}	17.88^{cd}	2.86^{b}	4.74^{b}	4.34^{b}
	180	15.78^{bcd}	38.57^{abc}	71.65^{bc}	60.30^{bcd}	17.62^{de}	2.89^{b}	4.79^{bc}	4.37^{b}
	270	16.03^{d}	38.97^{bc}	71.55^{bc}	60.05^{cd}	17.41^{de}	2.93^{b}	4.88^{bc}	4.43^{b}
	365	16.26^{d}	39.29^{c}	71.41^{c}	59.77^{d}	17.23^{e}	2.97^{b}	4.98^{c}	4.54^{c}
MSE		0.190	0.690	0.169	0.211	0.134	0.010	0.026	0.005
MSD		0.009	0.085	0.012	0.041	0.040	0.003	0.003	0.001

Means with different superscript letter in the same column of each factor were significantly different ($P < 0.05$)

MSE mean square error, *MSD* minimum significant difference, *FF* full-fat control (4.3%, w /v) cheese, *RF1* reduced fat (2.5%, w /v) cheese, *RF2* reduced fat (1.4%, w /v) cheese, *Culture M* MT1, *Culture MA* MT1 + LH-B02, *FDM* fat in dry matter, *MNFS* moisture in nonfat dry substance, *S/M* salt in moisture

Statistical analysis showed that the fat content was affected significantly ($P<0.05$) by ripening time. The reduction of fat significantly affected ($P<0.05$) the chemical composition of white, brined, goat cheese. More specifically, moisture, protein and salt content increased with reduction in fat, whereas fat in dry matter (% FDM) and moisture in non-fat substance (% MNFS) decreased. The protein and moisture content were the highest in the early stages of the ripening period for all cheese treatments, whereas they decreased as ripening proceeded. In contrast, the fat, salt and salt in moisture content increased with ripening. Salt in moisture in all samples was not affected ($P>0.05$) by reduction in fat. These results are similar to those reported for other low-fat cheeses such as feta type cheeses [24, 25, 35], low-fat, white, brined cheeses [36, 37], reduced and low-fat, artisanal-style goat cheese [38] and Cheddar cheese [21].

Salt content is higher in the low-fat cheeses due to lower fat or higher protein content which contributes to more binding sites for salt and can increase its retention in the curd [35]. Moisture in non-fat substance was higher in full-fat cheeses compared to other treatments. After day 30 of ripening there were no significant differences in MNFS% in all cheese samples. Such a behavior has been also observed for low-fat feta-type cheese [24, 35]. The variation in moisture of white, brined cheeses during ripening is due to the cleavage of peptide bonds, the result of proteolysis. As peptide bonds are cleaved, two new ionic groups are generated and each of these will compete for the available water in the system. Thus, part of the water previously available will be chemically bound to these new ionic groups and will not be determined by the classical oven drying method [39].

The use of the adjunct culture significantly affected only protein content ($P<0.05$). Present results are in agreement with studies in the literature involving the addition of adjunct cultures in white, brined cheeses [24, 31, 40].

The pH values of all samples were similar to those found in other relevant studies dealing with white, brined cheeses produced with goat milk [31, 39, 41, 42]. Higher ($P<0.05$) pH values were observed for reduced-fat (RF2) samples while (RF1) and (FF) cheese samples did not differ significantly ($P>0.05$). The use of the adjunct culture did not affect pH values. The pH values of all samples decreased during the first stage of ripening (until samples were transferred to the cold room) due to the loss of moisture [31] and the production of lactic acid by the starter culture. This stage is very important to avoid defects since the rapid and adequate production of lactic acid is crucial for brined cheeses [43]. The increase in pH of all samples after the second stage of ripening is caused by utilization of lactic acid, formation of non-acidic decomposition products and liberation of protein decomposition alkaline products [44]. The pH of cheese is influenced by salt content since salt can affect microbial growth. Thus, the activity of lactic acid bacteria and non-starter lactic acid bacteria depends on the salt content and especially for white, brined cheeses on the salt in moisture content [43, 44].

Proteolysis

Proteolysis is the principal and most complex biochemical event which occurs during the ripening of most cheese varieties. It is desirable for low-fat cheese starters to have low proteolytic but high peptidolytic activities [11]. In the present study the degree of proteolysis (Table 3) was evaluated by measuring (i) the % WSN which is due to the formation of small and medium sized peptides and free amino acids, (ii) the % TCA-SN including free amino acids, small- and medium-size peptides, and smaller nitrogen compounds, such as amines, urea and ammonium and (iii) the % PTA–SN including free amino acids and very small peptides amino acids and smaller Nitrogen compounds (except dibasic amino acids [Arg, Cit, Lys] and ammonia) [7, 14].

All nitrogen fractions of cheese samples were markedly affected ($P<0.05$) by the reduction of fat, the use of adjunct

Table 3 Evolution of nitrogen fractions of full- and reduced-fat, white, brined goat cheese with or without adjunct culture during aging, analyzed by ANOVA

Factors		TN	% WSN	% TCA-SN	% PTA-SN
Treatment	FF	2.61^{a}	10.54^{a}	7.76^{a}	3.54^{a}
($n=64$)	RF1	2.74^{b}	8.87^{b}	6.21^{b}	2.66^{b}
	RF2	3.09^{c}	6.94^{c}	5.15^{c}	1.74^{c}
MSE		0.003	0.452	0.180	0.093
MSD		0.137	1.671	1.063	0.873
Culture	M	2.85^{a}	8.29^{a}	6.01^{a}	2.57^{a}
(n = 96)	MA	2.78^{b}	9.27^{b}	6.74^{b}	2.73^{b}
MSE		0.003	0.452	0.180	0.093
MSD		0.074	0.981	0.726	0.163
Days	1	2.96^{a}	5.76^{a}	3.11^{a}	1.06^{a}
($n=24$)	9 – 11	2.91^{ab}	6.91^{b}	4.65^{b}	1.70^{b}
	30	2.84^{bc}	7.33^{bc}	5.45^{c}	1.97^{b}
	60	2.80^{cd}	8.05^{c}	5.91^{c}	2.39^{c}
	120	2.80^{cd}	9.06^{d}	6.78^{d}	2.84^{d}
	180	2.76^{de}	9.79^{d}	7.48^{e}	3.31^{e}
	270	2.73^{de}	11.18^{e}	8.24^{f}	3.71^{f}
	365	2.70^{e}	12.16^{f}	9.37^{g}	4.20^{g}
MSE		0.003	0.452	0.180	0.093
MSD		0.004	0.421	0.461	0.268

Means with different superscript letter in the same column of each factor were significant different ($P<0.05$)

MSE mean square error, *MSD* minimum significant difference, *FF* full-fat control (4.3%, w /v) cheese, *RF1* reduced fat (2.5%, w /v) cheese, *RF2* reduced fat (1.4%, w /v) cheese, *Culture M* MT1, *Culture MA* MT1 + LH-B02, *TN* total nitrogen, *WSN* water soluble nitrogen, *TCA-SN* nitrogen soluble in 12% TCA, *PTA-SN* nitrogen soluble in 5% phosphotungistic acid

cultures and ripening time in the same way as protein content. Fat reduction raised TN of cheeses while lower TN values were observed for samples containing the adjunct culture. A statistically significant reduction ($P<0.05$) in TN was recorded during ripening, since a part of the nitrogen content migrates to the brine. These findings are in good agreement with reported data on white, full-fat, brined cheeses made from goat milk [5, 41, 42, 45].

It can also be observed that the content of the % WSN, % TCA-SN and % PTA-SN increased during ripening ($P<0.05$). These findings are in agreement with other studies [13, 35, 36, 40–42, 45–48]. As expected, the formation of % WSN, % TCA-SN and % PTA-SN was higher in full fat cheeses compared to reduced-fat counterparts. These results are in agreement with those for other white, brined, low-fat cheeses [35, 48]. The adjunct culture significantly increased ($P<0.05$) the values of all fractions % WSN, % TCA-SN and % PTA-SN. Tungjaroenchai et al. [49] reported the highest total free amino acid (FAA) content in reduced fat Edam cheese samples containing *Lactobacillus helveticus* adjunct culture, due to this culture's higher proteolytic activity compared to other lactobacilli strains in fermented milks. The present study showed lower levels of soluble nitrogen in full-fat cheese samples compared to other full-fat, white, brined goat cheeses prepared using mesophilic [39] and thermophilic starters [41]. In contrast, these values were higher compared to those for goat teleme cheese [45]. These differences are probably attributed to differences in milk composition, the cheese making process, starter culture and salting conditions. Other studies on white, brined cheeses [45, 50] reported that ewe and goat cheeses have higher levels of % WSN than cheeses made using cow milk.

The activity of residual chymosin is high in white, brined cheeses due to the characteristics of these cheeses such as low pH, high moisture and particular cheese making process without heat treatment of the curd [51]. Low proteolysis of goat cheeses can be attributed to the inaccessibility of enzymes from rennin, plasmin, lactic acid bacteria and non-starter lactic acid bacteria to hydrolyze specific bonds of caseins, or to the genetic variants of goat milk [45]. Salt content and the method of salting play a decisive role in the degree of proteolysis. More specifically, high salt content and low moisture content can reduce the proteolytic activity in cheeses [52].

Lipolysis

The concentrations of free fatty acids (FA) for all cheese samples during ripening and storage are presented in Table 4. All fatty acids increased during the above period because of lipolysis ($P<0.05$), except of hexanoic acid ($P>0.05$). The most abundant fatty acids in all cheese samples independent of adjunct culture were acetic, palmitic and stearic acid recorded throughout storage. This tendency has also been reported by other researchers for various brined cheeses [53–58] and non-brined goat cheeses [59–61]. Although acetic acid was the dominant acid, being characteristic of brined cheeses [23, 53, 54], it is not considered as a fatty acid since it is produced from lactose fermentation (via pyruvate) and amino acid degradation by the cheese microflora [62, 63].

In general, fat reduction decreases the concentration of all fatty acids ($P<0.05$) except for acetic, hexanoic and stearic acid. Reduced fat RF2 cheese samples with adjunct culture had higher concentration of acetic acid over 120 days, compared to the FF cheese with the main culture only ($P<0.05$). The additional amount of acetic acid can be attributed to the peptidolytic activity of adjunct LH-B02 culture. Similar results were reported for low-fat feta type cheese made by ewe milk [53]. Angelopoulou et al. [63] reported that 60 days' ripened feta cheese inoculated with propionic adjunct culture enhanced the production of acetic acid. The adjunct culture (LH-B02) contributed ($P<0.05$) to the increase in butyric acid in mature (over120 days), reduced-fat cheeses samples of the same fat content. Butyric acid, regardless of the fat content and the culture used, showed the lowest concentration of all FAs. Such a finding was also reported for Xynotyri goat cheese [64], for saturated FA of goat cheese [61] and Rocamadour goat cheese [65]. According to Barlowska et al. [66] butyric acid is associated with bitterness as well as brine and goat milk odor. Hexanoic acid content was, also, not affected ($P>0.05$) by starter culture and fat content, as also observed by others [53]. Free Fatty acids caprylic, capric, myristic and palmitic acids were affected by fat reduction and were found in higher concentrations in full-fat samples compared to those in reduced-fat samples (RF1 and RF2) ($P<0.05$). McCarthy et al. [67] reported a similar behavior for caprylic acid, myristic acid and palmitic acid in reduced-fat and low-fat cheddar cheese samples.

The mean concentrations of short-chain ($C_{4:0}$–$C_{8:0}$), medium-chain ($C_{10:0}$–$C_{14:0}$), long-chain ($C_{16:0}$–$C_{18:0}$) and total free fatty acids of all cheese samples during ripening and storage are presented in Fig. 1. During ripening (1–60 days), short-chain FFAs did not differ significantly ($P>0.05$) among treatments. Hexanoic, lauric, myristic and palmitic acids were the main short-chain ($C_{4:0}$–$C_{8:0}$), medium-chain ($C_{10:0}$–$C_{14:0}$) and long-chain ($C_{16:0}$–$C_{18:0}$) FFAs, respectively, in all samples, irrespective of fat content and the use of adjunct culture. The sum of FFAs ($C_{4:0}$–$C_{18:0}$) for all cheeses, especially for full-fat ones, was similar to that found for goat Teleme cheese [54], but higher than that of artisanal goat cheese made from raw goat milk [38], and lower compared to the total amount of free fatty acids determined for white brined cheeses [23, 56, 57, 68, 69] or other non-brined goat cheeses [64, 65, 70, 71]. Differences

Table 4 Concentration of free fatty acids (mg/100 g) of full and reduced fat goat white brined cheese with or without adjunct culture

FFA	Age (days)	FF M	FF MA	RF1 M	RF1 MA	RF2 M	RF2 MA
C2:0	1	32.40 ± 3.54 aA	39.00 ± 2.97 aA	36.85 ± 2.90 aA	39.90 ± 2.83 aA	39.65 ± 4.03 aA	46.10 ± 5.52 aA
	9–11	48.25 ± 2.47 aA	57.00 ± 4.81 bAB	58.75 ± 4.60 bAB	64.00 ± 4.53 bAB	62.00 ± 4.53 bAB	67.00 ± 3.25 bB
	60	84.30 ± 4.81 bA	85.60 ± 4.94 cA	87.80 ± 2.69 cA	95.00 ± 5.09 cA	91.00 ± 5.37 cA	96.35 ± 3.89 cA
	120	95.40 ± 4.95 bA	98.00 ± 4.38 cdA	97.50 ± 4.10 cdA	101.50 ± 3.96 cA	98.60 ± 4.81 cA	104.25 ± 4.60 cdA
	180	100.95 ± 5.80 bA	105.15 ± 4.60 dAB	108.95 ± 4.31 dAB	111.00 ± 4.53 cAB	107.90 ± 4.38 cdAB	120.05 ± 4.74 dB
	365	120.50 ± 3.54 cA	127.75 ± 3.18 eAB	123.00 ± 1.41 eA	132.15 ± 3.61 dAB	125.25 ± 3.89 dAB	138.00 ± 4.53 eB
C4:0	1	0.58 ± 0.08 aAB	0.70 ± 0.08 aB	0.49 ± 0.02 aAB	0.63 ± 0.06 aAB	0.43 ± 0.03 aA	0.44 ± 0.04 aA
	9–11	0.62 ± 0.05 aABC	0.78 ± 0.05 aA	0.52 ± 0.05 aBC	0.71 ± 0.07 aAB	0.42 ± 0.02 aC	0.51 ± 0.11 aBC
	60	1.04 ± 0.09 bAB	1.18 ± 0.08 bA	0.86 ± 0.17 abAB	1.04 ± 0.12 bAB	0.67 ± 0.10 abB	0.90 ± 0.04 bAB
	120	1.32 ± 0.07 bcA	1.44 ± 0.05 bcA	0.84 ± 0.05 abB	1.38 ± 0.04 cA	0.73 ± 0.10 bcB	1.34 ± 0.04 cA
	180	1.47 ± 0.07 cA	1.61 ± 0.08 cA	0.92 ± 0.04 abB	1.60 ± 0.04 cA	0.83 ± 0.10 bcB	1.49 ± 0.11 cA
	365	1.87 ± 0.12 dA	2.01 ± 0.03 dA	1.22 ± 0.30 bB	1.98 ± 0.05 dA	1.00 ± 0.04 cB	1.97 ± 0.06 dA
C6:0	1	4.65 ± 0.63aA	4.81 ± 0.48aA	4.46 ± 0.34aA	4.61 ± 0.37aA	4.43 ± 0.38aA	4.61 ± 0.57aA
	9–11	4.83 ± 0.31aA	5.12 ± 0.42aA	4.58 ± 0.36aA	4.72 ± 0.44aA	4.49 ± 0.32aA	4.76 ± 0.60aA
	60	5.46 ± 0.31aA	5.63 ± 0.33aA	4.77 ± 0.27aA	5.12 ± 0.33aA	5.02 ± 0.21aA	5.06 ± 0.33aA
	120	5.72 ± 0.30aA	5.95 ± 0.46aA	4.95 ± 0.62aA	5.48 ± 0.53aA	5.10 ± 0.39aA	5.33 ± 0.27aA
	180	5.79 ± 0.20aA	6.28 ± 0.54aA	5.26 ± 0.46aA	5.64 ± 0.28aA	5.11 ± 0.49aA	5.36 ± 0.39aA
	365	6.25 ± 0.47aA	6.64 ± 0.60aA	5.88 ± 0.55aA	6.05 ± 0.65aA	5.47 ± 0.32aA	6.17 ± 0.34aA
C8:0	1	0.71 ± 0.04 aAB	0.78 ± 0.10 aB	0.58 ± 0.03 aAB	0.76 ± 0.04 aAB	0.51 ± 0.07 aA	0.69 ± 0.06 aAB
	9–11	1.08 ± 0.03 aA	1.19 ± 0.14 aA	0.82 ± 0.03 aAB	0.95 ± 0.17 aAB	0.63 ± 0.08 abB	0.77 ± 0.12 abAB
	60	1.93 ± 0.13 bA	1.98 ± 0.13 bA	1.41 ± 0.12 bB	1.56 ± 0.11 cAB	1.07 ± 0.11 bcB	1.22 ± 0.15 bcB
	120	2.36 ± 0.13 bcA	2.47 ± 0.20 bA	1.57 ± 0.05 bB	1.69 ± 0.16 cB	1.27 ± 0.18 cdB	1.46 ± 0.11 cdB
	180	3.00 ± 0.27 cA	3.24 ± 0.10 cA	2.15 ± 0.10 cB	2.26 ± 0.19 dB	1.74 ± 0.18 dB	1.85 ± 0.12 dB
	365	4.69 ± 0.25 dA	4.70 ± 0.31 dA	3.19 ± 0.17 dB	3.37 ± 0.12 eB	2.39 ± 0.17 eC	2.57 ± 0.11 eBC
C10:0	1	2.63 ± 0.25 aA	2.55 ± 0.45 aA	1.32 ± 0.23 aB	1.43 ± 0.28 aB	0.92 ± 0.11 aB	1.01 ± 0.21 aB
	9–11	2.80 ± 0.24 aA	2.98 ± 0.13 abA	1.63 ± 0.17 abB	1.86 ± 0.18 abB	1.24 ± 0.32 aB	1.55 ± 0.20 aB
	60	4.02 ± 0.41 bA	4.28 ± 0.26 bcA	2.22 ± 0.27 abB	2.43 ± 0.19 abB	1.88 ± 0.49 abB	2.03 ± 0.35 abB
	120	4.36 ± 0.39 bA	4.66 ± 0.54 cdA	2.68 ± 0.62 abcBC	2.75 ± 0.58 abABC	1.92 ± 0.24 abC	2.16 ± 0.41 abC
	180	5.57 ± 0.19 cA	5.88 ± 0.25 dA	3.03 ± 0.51 bcB	3.25 ± 0.48 bB	2.68 ± 0.45 bcB	3.04 ± 0.43 bcB
	365	7.59 ± 0.04 dA	7.63 ± 0.18 eA	4.17 ± 0.33 cBC	4.82 ± 0.25 cB	3.46 ± 0.13 cC	3.58 ± 0.20 cC
C12:0	1	4.39 ± 0.70 aAB	5.04 ± 0.33 aA	3.32 ± 0.32 aBC	3.83 ± 0.25 aAB	2.28 ± 0.31 aC	3.14 ± 0.22 aBC
	9–11	4.62 ± 0.38 aAB	5.57 ± 0.40 aA	3.52 ± 0.52 abBC	4.32 ± 0.46 aABC	2.68 ± 0.40 abC	3.85 ± 0.30 abBC
	60	6.54 ± 0.48 bA	8.36 ± 0.36 bB	4.94 ± 0.38 bcAC	6.44 ± 0.42 bA	4.14 ± 0.41 bcC	5.47 ± 0.38 bcAC
	120	8.92 ± 0.29 cAB	10.26 ± 0.31 cA	6.14 ± 0.42 cdCD	7.77 ± 0.48 bcBC	4.95 ± 0.46 cdD	6.95 ± 0.75 cdC
	180	10.33 ± 0.41 cdAB	10.81 ± 0.39 cA	6.96 ± 0.31 deCD	8.51 ± 0.32 cdBC	5.60 ± 0.61 cdD	7.88 ± 0.74 dD
	365	10.96 ± 0.46 dA	11.69 ± 0.40 cA	7.90 ± 0.39 eB	9.31 ± 0.31 dB	6.12 ± 0.42 dC	8.47 ± 0.44 dB
C14:0	1	6.35 ± 0.53 aAB	6.81 ± 0.35 aA	4.11 ± 0.60 aC	4.54 ± 0.46 aBC	2.62 ± 0.37 aC	3.43 ± 0.56 aC
	9–11	6.97 ± 0.46 abA	7.76 ± 0.33 aA	4.83 ± 0.32 abB	5.24 ± 0.41 abB	3.03 ± 0.33 abC	3.89 ± 0.35 aBC
	60	7.98 ± 0.50 abAB	8.51 ± 0.41 abA	5.34 ± 0.53 abcCD	6.58 ± 0.37 bcBC	4.19 ± 0.29 bcD	4.66 ± 0.50 abD
	120	8.90 ± 0.47 bcAB	10.37 ± 0.83 bcA	6.41 ± 0.42 bcdCD	7.15 ± 0.30 cdBC	4.76 ± 0.38 cdD	5.89 ± 0.45 bcCD
	180	10.48 ± 0.58 cdA	11.20 ± 0.48 cA	6.75 ± 0.46 cdBC	7.62 ± 0.29 cdB	5.41 ± 0.43 cdC	6.45 ± 0.25 cBC
	365	11.19 ± 0.54 dA	11.86 ± 0.67 cA	7.96 ± 0.45 dBC	8.56 ± 0.42 dB	6.15 ± 0.41 dC	7.30 ± 0.37 cBC
C16:0	1	8.82 ± 1.02 aAB	9.65 ± 0.81 aA	5.99 ± 1.17 aABC	5.96 ± 1.39 aABC	3.82 ± 0.97 aC	4.82 ± 0.67 aBC
	9–11	11.91 ± 1.17 abAB	12.73 ± 0.93 abA	7.64 ± 2.01 abBC	10.45 ± 1.46 abABC	5.61 ± 0.75 abC	6.88 ± 0.65 aC
	60	14.04 ± 0.93 bcAB	17.72 ± 0.68 bcA	11.38 ± 1.44 abcBC	14.01 ± 1.47 bAB	7.21 ± 1.28 abC	8.30 ± 1.06 abC
	120	15.86 ± 0.41 cAB	20.48 ± 1.58 cA	13.45 ± 0.88 bcBC	15.78 ± 0.98 bcB	10.05 ± 1.35 bcC	12.28 ± 1.46 bcBC
	180	23.23 ± 0.91 dA	26.01 ± 1.52 dA	17.04 ± 1.87 cBC	20.92 ± 1.64 cdAB	12.38 ± 0.80 cC	14.31 ± 1.12 cdC
	365	27.02 ± 1.15 dA	29.31 ± 2.01 dA	23.10 ± 1.42 dAB	25.98 ± 1.42 dA	17.46 ± 1.81 dB	19.10 ± 1.84 dB

Table 4 (continued)

FFA	Age (days)	FF M	FF MA	RF1 M	RF1 MA	RF2 M	RF2 MA
C18:0	1	7.37 ± 0.93 aAB	8.20 ± 0.68 aB	5.82 ± 0.92 aAB	6.21 ± 1.51 aAB	3.74 ± 0.89 aA	4.38 ± 1.15 aAB
	9–11	7.82 ± 0.91 aA	8.60 ± 0.74 aA	7.37 ± 1.07 aA	7.49 ± 1.51 abA	4.20 ± 1.25 abA	4.87 ± 1.09 aA
	60	9.82 ± 0.64 abA	9.97 ± 0.96 abA	8.02 ± 1.04 abAB	8.43 ± 0.92 abAB	5.25 ± 0.70 abB	5.58 ± 1.05 aB
	120	10.73 ± 1.43 abAB	13.33 ± 1.09 abcB	9.65 ± 1.23 abAB	11.91 ± 1.46 abcB	5.86 ± 1.13 abA	6.20 ± 1.16 aA
	180	12.22 ± 1.72 abAB	15.09 ± 1.92 bcA	11.23 ± 2.05 abAB	13.42 ± 1.70 bcAB	7.84 ± 1.47 abB	8.65 ± 1.60 abAB
	365	15.26 ± 2.09 bAB	17.75 ± 2.30 cB	13.48 ± 1.81 bAB	16.40 ± 1.79 cAB	9.37 ± 1.96 bA	12.04 ± 1.91 bAB

Data are expressed as means ± standard deviation. Means with different lower case superscript letter in the same column are significantly different ($P < 0.05$). Means with different capital superscript number in the same row are significant different ($P < 0.05$)

FF full-fat control (4.3%, w /v) cheese, *RF1* reduced fat (2.5%, w /v) cheese, *RF2* reduced fat (1.4%, w /v) cheese, *Culture M* MT1, *Culture MA* MT1 + LH-B02

in total FFAs in the present study vs. those in the literature may be due to the low lipolytic activity of the main culture in combination with the short period of the first ripening or even different kind of cheese making process. Other factors that can affect the FFA profile are the herd diet and lactation stage [65]. Total FFAs increased during ripening and storage ($P < 0.05$) and were significantly affected by fat reduction especially for mature cheese samples ($P < 0.05$). For a given fat content there were statistically significant differences in FFA concentration between different culture treatments of mature cheese samples.

Texture profile analysis

Texture evaluation of reduced fat, white, brined goat cheese prepared with or without an adjunct culture as a function of ripening time is shown in Fig. 2. Statistical analysis of the cheese rheological properties indicated that the reduced-fat cheeses were harder and less brittle, showing increased force at the point of fracture, compared to full-fat cheeses ($P < 0.05$). These findings are in agreement with the results reported for low-fat, feta type cheese [72], low-fat, white, brined cheeses [36, 37] and other types of low- and reduced-fat cheeses [20, 21, 38, 52, 73]. Increased hardness and fracture stress of low-fat, white, brined cheeses is attributed to the more continuous protein matrix prevailing due the decreased fat content in low-fat cheeses [37, 72].

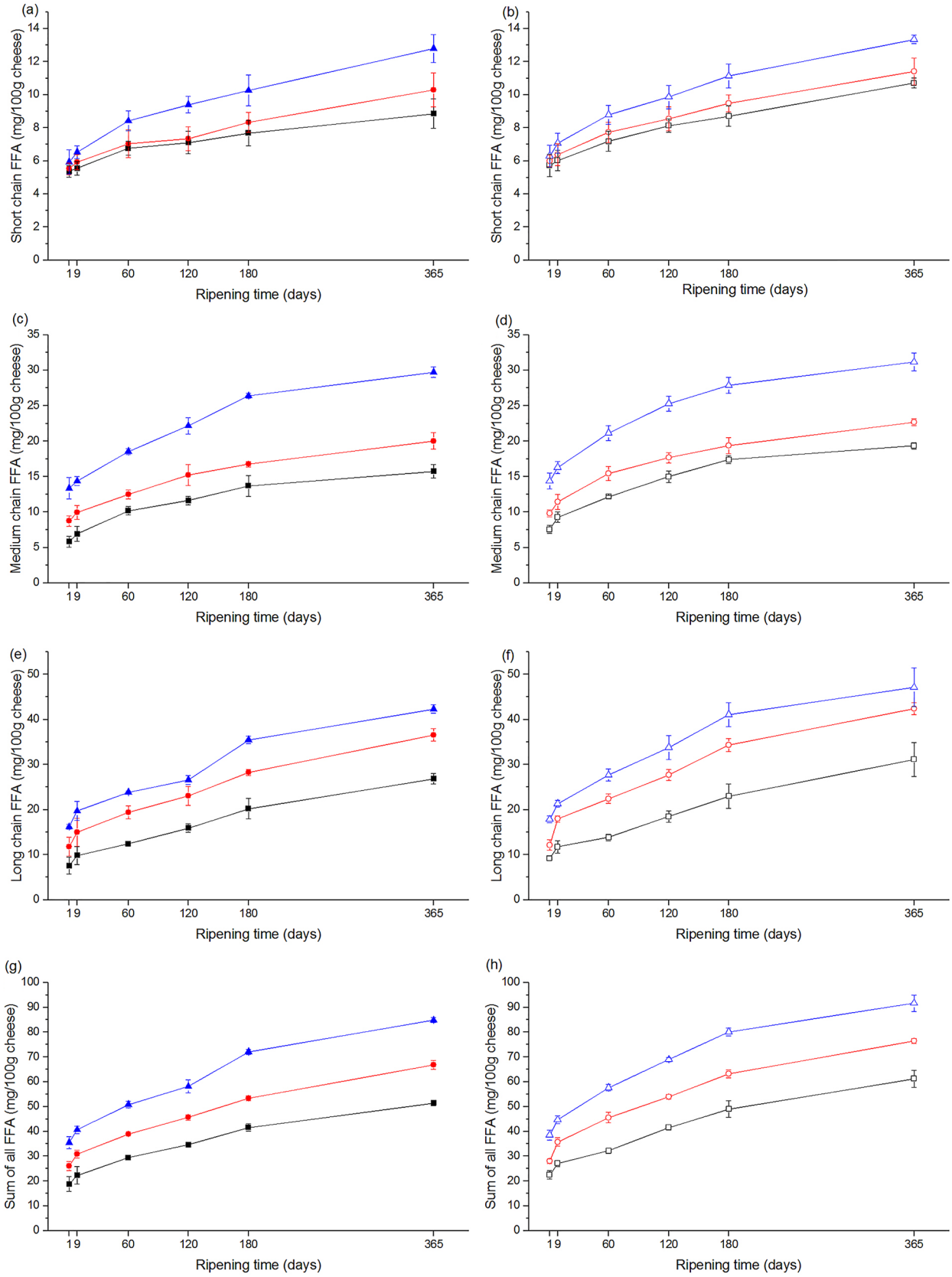
At the second stage of ripening (9–11 to 60 day), hardness and force at the point of fracture increased. At the later stages of ripening (after 60 days) all the texture parameters decreased. This is most likely attributed to the enhanced proteolysis during ripening [24, 35–37, 74].

The use of the (MT1) mesophilic/ thermophilic and thermophilic (adjunct) (LH-B02) cultures in the present study led to the decrease in values ($P < 0.05$) of all textural properties. Similar results were reported for feta type cheese [75] and teleme cheese [74]. According to Mistry [11] and Karami et al. [76], starter cultures play an important role in cheese quality due to their contribution to proteolysis, texture and flavor development through slow but progressive breakdown of caseins during ripening. Tsigkros et al. [77] observed that feta cheese made using 30% of goat milk was harder than that made with ovine milk. Mallatou et al. [42] and Pappa et al. [74] noted that goat milk cheeses required a higher force to fracture compared to cheeses made from other types of milk. This may be probably attributed to the different casein structure and lower casein content of goat milk compared to that of ewe milk. In addition, the network structure of cheese is affected by factors such as chemical composition, degree of proteolysis, fat droplet size and distribution as well as casein–casein, casein–water and casein–fat interactions, the state of water (bulk or bound to casein matrix), pH and the state of calcium (ionic or bound to casein matrix) [78].

Sensory evaluation

Sensory evaluation results (appearance, body/texture, flavor and total score), as a function of ripening time are presented in Fig. 3. Ripening time did not affect ($P > 0.05$) body/texture, flavor and sum of all sensory attributes. This is very important since cheeses of up to 1 year may be consumed without exhibiting over-ripening problems. Bitterness and off-flavors are common defects in aged, low-fat cheeses, which are usually developed early in the ripening process due to the low salt content, high moisture and lower proportion of aromatic

Fig. 1 Concentrations of short-chain ($C_{4:0}$–$C_{8:0}$), medium-chain ($C_{10:0}$–$C_{14:0}$), long-chain ($C_{16:0}$–$C_{18:0}$) and total fatty acids of all cheese samples during ripening and storage. Full-fat control (4.3%, w /v) with MT1 culture (FF M) (▲), Reduced-fat (2.5%, w /v) cheese with MT1 culture (RF1 M) (●), Reduced-fat (1.4%, w /v) cheese with MT1 culture (RF2 M) (■), Full-fat control (4.3%, w /v) with MT1 + LH-B02 culture (FF MA) (Δ), Reduced-fat (2.5%, w /v) cheese with MT1 + LH-B02 (RF1 MA) (○), Reduced-fat (1.4%, w /v) cheese with MT1 + LH-B02 (RF2 MA) (□) ▶


(a)
Short chain FFA (mg/100g cheese)
Ripening time (days)
(b)
Short chain FFA (mg/100g cheese)
Ripening time (days)
(c)
Medium chain FFA (mg/100g cheese)
Ripening time (days)
(d)
Medium chain FFA (mg/100g cheese)
Ripening time (days)
(e)
Long chain FFA (mg/100g cheese)
Ripening time (days)
(f)
Long chain FFA (mg/100g cheese)
Ripening time (days)
(g)
Sum of all FFA (mg/100g cheese)
Ripening time (days)
(h)
Sum of all FFA (mg/100g cheese)
Ripening time (days)
1 9
60
120
180
365

compounds formed through fat degradation [11]. According to Sousa et al. [15] the hydrophobic bitter peptides in full-fat cheeses are not easily detected by sensory panelists due to partitioning of such peptides into the fat phase. Salt can affect the production of bitter peptides by promoting hydrophobic interactions between susceptible regions of β-Casein, thus reducing chymosin action on protein. In the present study, no bitterness and off-flavors were detected by panelists in all samples during the entire ripening period probably due to the specific salt content and low pH values.

The panelists detected differences in appearance, body-texture and flavor ($P < 0.05$) between full-fat and reduced-fat cheeses. They found reduced-fat cheeses to be harder than the full-fat samples, although well accepted by panelists over a 12-month period. In general, panelists showed a marked preference to samples made with the adjunct culture. It should be noted that in a previous study panelists showed preference for Prato cheese made using the adjunct culture *Lactobacillus helveticus B02* [46].

Fig. 3 Sensory evaluation of full and reduced fat, white, brined goat cheese made with or without the addition of adjunct culture during aging. FF: Full-fat control (4.3%, w /v) cheese, RF1: Reduced-fat (2.5%, w /v) cheese, RF2: Reduced-fat (1.4%, w /v) cheese. Culture: MT1 (M) or MT1 + LH-B02 (MA) ▶

Pearson's correlation

Correlation between chemical composition, pH, proteolysis, texture properties and sensory attributes of all cheese samples was attempted after 4 months of cheese ripening (on day 120) (Table 5). A significant negative correlation was obtained between texture properties such as hardness and force at the point of fracture with % WSN, % TCA-SN,

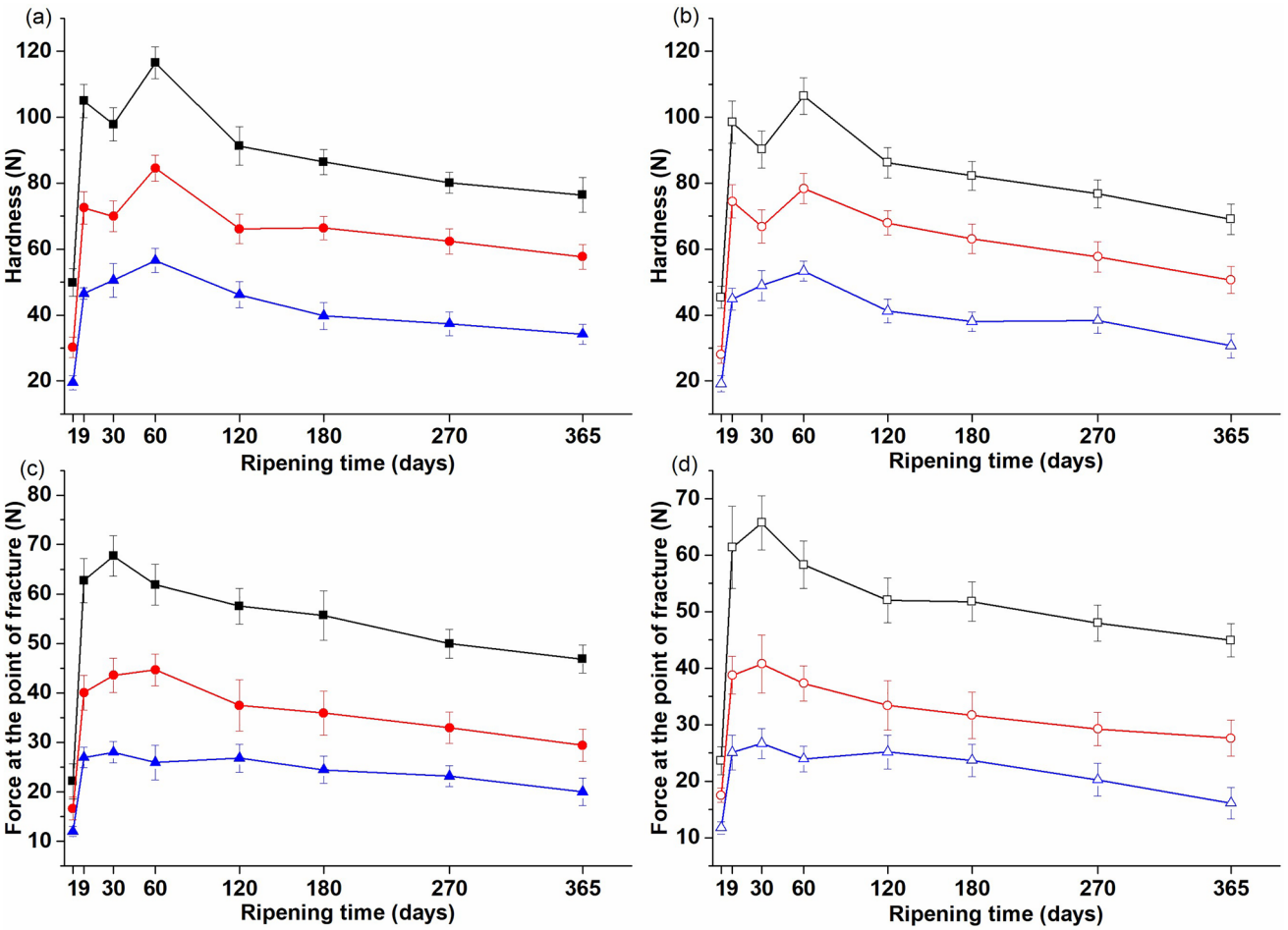


Fig. 2 Hardness (**a** and **b**) and force at the point of fracture (**c** and **d**) of full- and reduced-fat, white, brined goat cheese made with or without the addition of adjunct culture during aging. Full-fat control (4.3%, w /v) with MT1 culture (FF M) (▲), Reduced-fat (2.5%, w /v) cheese with MT1 culture (RF1 M) (●), Reduced-fat (1.4%, w /v) cheese with MT1 culture (RF2 M) (■), Full-fat control (4.3%, w /v) with MT1 + LH-B02 culture (FF MA) (Δ), Reduced-fat (2.5%, w /v) cheese with MT1 + LH-B02 (RF1 MA) (○), Reduced-fat (1.4%, w /v) cheese with MT1 + LH-B02 (RF2 MA) (□)

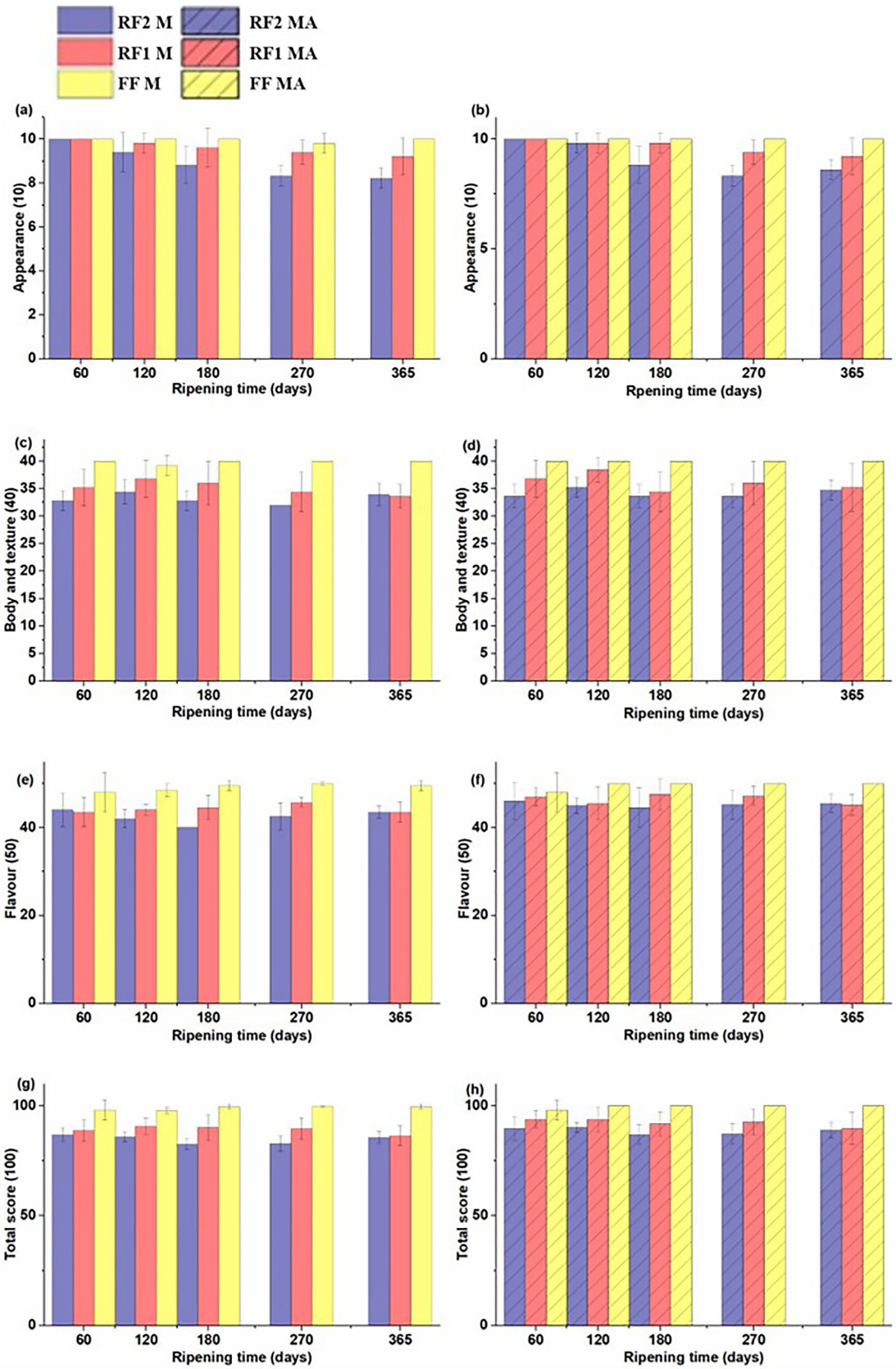

RF2 M
RF2 MA
RF1 M
RF1 MA
FF M
FF MA
(a)
Appearance (10)
Ripening time (days)
(b)
Appearance (10)
Rpening time (days)
(c)
Body and texture (40)
Ripening time (days)
(d)
Body and texture (40)
Ripening time (days)
(e)
Flavour (50)
Ripening time (days)
(f)
Flavour (50)
Ripening time (days)
(g)
Total score (100)
Ripening time (days)
(h)
Total score (100)
Ripening time (days)
60
120
180
270
365

Table 5 Correlation between chemical composition, pH, texture parameters and sensory attributes of goat cheeses on day 120

	Moisture	Fat	pH	Salt	TN	% WSN	% TCA-SN	% PTA-SN	Hardness (*N*)	FF (*N*)	Body and texture
Fat	−0.998**										
pH	0.510	−0.514									
Salt	0.962**	−0.961**	0.685*								
TN	0.842**	−0.858**	0.729**	0.846**							
% WSN	−0.845**	0.847**	−0.680*	−0.832**	−0.950**						
% TCA-SN	−0.832**	0.846**	−0.529	−0.810**	−0.880**	0.893**					
% PTA-SN	−0.951**	0.961**	−0.567	−0.927**	−0.895**	0.870**	0.871**				
Hardness (*N*)	0.943**	−0.957**	0.575	0.918**	0.921**	−0.867**	−0.865**	−0.989**			
FF (*N*)	0.862**	−0.877**	0.669*	0.851**	0.960**	−0.916**	−0.846**	−0.949**	0.958**		
Body and texture	−0.758**	0.755**	−0.549	0.731**	0.800**	−0.839**	−0.716**	−0.678*	−0.684*	−0.660*	
Flavor	−0.805**	0.804**	−0.338	−0.723**	−0.747**	0.750**	0.787**	0.797**	−0.540	−0.531	0.685*

FF force at the point of fracture, *TN* total nitrogen, *WSN* water soluble nitrogen, *TCA-SN* nitrogen soluble in 12% TCA, *PTA-SN* nitrogen soluble in 5% phosphotungistic acid

*Correlation is significant at the 0.05 level (2-tailed)

**Correlation is significant at the 0.01 level (2-tailed)

% PTA-SN and fat content. Pappa et al. [74] reported that goat teleme cheese had higher hardness values correlated to lower values of % WSN and % TCA–SN. Body and texture were found to have a significant negative correlation with moisture, % WSN, % TCA-SN, % PTA-SN and instrumental texture properties, while fat, salt and TN had a significant positive correlation. According to Romeih et al. [36] the reduction in values of texture parameters with increasing proteolysis is certainly a reflection of casein network disruption. This in turn, leads to the reorganization and the release of fat globules from the casein matrix [76]. On the contrary, a significant positive correlation was obtained between texture properties such as hardness and force at the point of fracture with moisture, salt and total nitrogen content. The salt content significantly affected all the texture parameters in the present study, as also observed in other studies [21, 44, 52]. Biochemical reactions are affected by moisture, pH and salt content since these parameters determine the type of bacteria and enzymic activities that may occur [7]. According to McCarthy et al. [21], fracture of hard cheeses such as Cheddar, is affected by casein, moisture, fat salt content and pH. Flavor had a significant positive correlation with fat and proteolysis. According to Molina et al. [50] and Tzanetakis et al. [39] the higher levels of Nitrogen soluble in phosphotungstic acid fraction acts as a precursor to other compounds essential for characteristic flavor development.

Conclusion

Overall, results of the present study indicate that the combination of mixed mesophilic/ thermophilic as main and thermophilic as adjunct starter culture can enhance proteolysis in reduced-fat, white, brined goat cheeses, resulting in the improvement of cheese sensory characteristics (flavor and texture). Therefore, our approach may be useful in producing white, brined, goat cheeses that will combine reduced-fat content and sensory attributes well acceptable by consumers.

Acknowledgements We are grateful to Dodoni S.A. for providing the milk and cultures and to the Dairy School of Ioannina for providing access to the cheese making equipment.

Declarations

Conflict of interest The authors declare no conflict of interest.

Compliance with ethics requirements This article does not contain any studies with human participants or animals performed by any of the authors.

Research involving human participants and/or animals The present study does not involve research on the use of human participants and/or animals.

References

1. Assis POA, Guerra GCB, Araújo DFS, Araújo JRFA, Machado TADG, Araújo AA, Lima TAS, Garcia HEM, Andrade LFLI, Queiroga RCRE (2016) Intestinal anti-inflammatory activity of goat milk and goat yoghurt in the acetic acid model of rat colitis. Int Dairy J 56:45–54
2. Hodgkinson AJ, Wallace OAM, Boggs I, Broadhurst M, Prosser CG (2018) Gastric digestion of cow and goat milk: impact of infant and young child in vitro digestion conditions. Food Chem 245:275–281
3. Kalyan S, Meena S, Kapila S, Sowmya K, Kumar R (2018) Evaluation of goat milk fat and goat milk casein fraction for anti-hypercholesterolaemic and antioxidative properties in hypercholesterolaemic rats. Int Dairy J 84:23–27
4. Verruck S, Dantas A, Schwinden Prudencio E (2019) Functionality of the components from goat's milk, recent advances for functional dairy products development and its implications on human health. J Funct Foods 52:243–257
5. Kocak A, Sanli T, Anlib EA, Hayaloglu AA (2020) Role of using adjunct cultures in release of bioactive peptides in white brined goat-milk cheese. LWT - Food Sci Technol 123:1–8
6. FAOSTAT (2021) Food and Agriculture Organization of the United Nations, Statistics Division. Data. http://www.fao.org/faostat/en/#data/QL. Accessed 26 Mar 2021
7. Moatsou G, Govaris A (2011) White brined cheeses: A diachronic exploitation of small ruminants milk in Greece. Small Rumin Res 101:113–121
8. Greek Foods and Beverages Codex (2014) https://www.aade.gr/sites/default/files/2020-03/83-iss3.pdf. Accessed 26 Mar 2021
9. Farkye NY, Guinee TP (2017) Low-fat and low-sodium cheeses. In: McSweeney PLH, Fox PF, Cotter PD, Everett DW (eds) Cheese chemistry, physics & microbiology, vol 1, 4th edn. Elsevier Academic Press, UK
10. Mohamed AG (2015) Low-fat cheese: a modern demand. Int J Dairy Sci 10(6):249–265
11. Mistry VV (2001) Low fat cheese technology. Int Dairy J 11(4–7):413–422
12. Baptista DP, Galli BD, Cavalheiro FG, Negrão F, Eberlin MN, Gigante ML (2018) Lactobacillus helveticus LH-B02 favours the release of bioactive peptide during Prato cheese ripening. Int Dairy J 87:75–83
13. Dimitrellou D, Kandylis P, Mallouchos A, Komaitis M, Koutinas A, Kourkoutas Y (2010) Effect of freeze-dried kefir culture on proteolysis in feta-type and whey-cheeses. Food Chem 119:795–800
14. Ardö Y, McSweeney PLH, Magboul AAA, Fox PF (2017). In: McSweeney PLH, Fox PF, Cotter PD, Everett DW (eds) Cheese chemistry, physics & microbiology, vol 1, 4th edn. Elsevier Academic Press, UK
15. Sousa MJ, Ardö Y, McSweeney PLH (2001) Advances in the study of proteolysis during cheese ripening. Int Dairy J 11(4–7):327–345
16. Thierry A, Collins YF, Abeijòn Mukdsi MC, Mc Sweeney PLH, Wilkinson MG, Spinnler HE (2017). In: McSweeney PLH, Fox PF, Cotter PD, Everett DW (eds) Cheese chemistry, physics & microbiology, vol 1, 4th edn. Elsevier Academic Press, UK
17. Ganesan B, Weimer BC (2017) Amino acid catabolism and its relationship to cheese flavor outcomes. In: McSweeney PLH, Fox PF, Cotter PD, Everett DW (eds) Cheese chemistry, physics & microbiology, vol 1, 4th edn. Elsevier Academic Press, UK
18. Smit G, Smit BA, Engels WJM (2005) Flavour formation by lactic acid bacteria and biochemical flavor profiling of cheese products. FEMS Microbiol Rev 29:591–610
19. Sanchez-Macias D, Morales-delaNuez A, Moreno-Indias I, Hernandez-Castellano LE, Mendoza-Grimon V, Castro N, Arguello A (2011) Lipolysis and proteolysis profiles of fresh artisanal goat cheese made with raw milk with 3 different fat contents. J Dairy Sci 94(12):5786–5793
20. Nateghi L, Roohinejad S, Totosaus A (2012) Physicochemical and textural properties of reduced fat Cheddar cheese formulated with xanthan gum and/or sodium caseinate as fat replacers. J Food Agric Environ 10(2):59–63
21. McCarthy CM, Wilkinson MG, Kelly PM, Guinee T (2016) Effect of salt and fat reduction on proteolysis, rheology and cooking properties of Cheddar cheese. Int Dairy J 56:74–86
22. Baghdadi F, Aminifar M, Farhoodi M, Ali Abadi SS (2017) Study of macromolecular interactions in low-fat brined cheese modified with Zedu gum. Int J Dairy Technol 71(2):1–13
23. Kondyli E, Pappa EC, Kremmyda A, Arapoglou D, Metafa M, Eliopoulos C, Israilides C (2020) Manufacture of reduced fat white-brined cheese with the addition of β-glucans biobased polysaccharides as textural properties improvements. Polymers 12(2647):1–16
24. Katsiari MC, Voutsinas LP, Kondyli E, Alichanidis E (2002) Flavour enhancement of low-fat feta-type cheese using a commercial adjunct culture. Food Chem 79(2):193–198
25. Michaelidou AM, Katsiari MC, Kondyli E, Voutsinas LP, Alichanidis E (2003) Effect of a commercial adjunct culture on proteolysis in low-fat feta-type cheese. Int Dairy J 13(2):179–189
26. FAO Food and Nutrition Paper (1986) In: Manuals of food quality control. 8. Food analysis: quality, adulteration and tests of identity. Rome
27. International Organization for Standardization (1975) Cheese Determination of fat content Gerber van Gulik method. ISO 3433. Geneva, Switzerland
28. International Dairy Federation (1982) Cheese and processed cheese: determination of the total solid content. IDF Standard 4A. Brussels, Belgium
29. Johnson ME, Olson NF (1985) A comparison of available methods for determining salt levels in cheese. J Dairy Sci 68:1020–1024
30. International Dairy Federation (1993) Milk Determination of nitrogen content. IDF Standard 20B. Brussels, Belgium
31. Pappa EC, Kandarakis I, Anifantakis EM, Zerfiridis GK (2006) Influence of types of milk and culture on the manufacturing practices, composition and sensory characteristics of teleme cheese during ripening. Food Control 17(7):570–581
32. De Jong C, Bandings HT (1990) Determination of free fatty acids in milk and cheese: procedures for extraction, clean up, and capillary gas chromatographic analysis. J High Resolut Chrom 13:94–98
33. Bourne MC (1978) Texture profile analysis. Food Technol 62:62–67
34. International Dairy Federation (1987) Sensory Evaluation of Dairy Products. IDF Standard 99A. Brussels, Belgium
35. Katsiari MC, Voutsinas LP (1994) Manufacture of low-fat feta cheese. Food Chem 49(1):53–60
36. Romeih EA, Michaelidou A, Biliaderis CG, Zerfiridis GK (2002) Low-fat white-brined cheese made from bovine milk and two commercial fat mimetics: chemical, physical and sensory attributes. Int Dairy J 12(6):525–540
37. Rahimi J, Khosrowshahi A, Madadlou A, Aziznia S (2007) Texture of low-fat Iranian white cheese as influenced by gum tragacanth as a fat replacer. J Dairy Sci 90(9):4058–4070
38. Sánchez-Macías D, Fresno M, Moreno-Indias I, Castro N, Morales-delaNuez A, Álvarez S, Argüello A (2010) Physicochemical analysis of full-fat, reduced-fat and low-fat artisan-style goat cheese. J Dairy Sci 93(9):3950–3956

39. Tzanetakis N, Vafopoulou – Mastrojiannaki A, Litopoulou – Tzanetaki E, (1995) The quality of white-brined cheese from goat's milk made with different starters. Food Microbiol 12:55–63
40. Tarakci Z, Tunctrurk Y (2008) The effect of adjunct cultures on some chemical and biochemical properties of white brined cheese. J Food Biochem 32(4):490–505
41. Kondyli E, Pappa EC, Svarnas C (2016) Ripening changes of the chemical composition, proteolysis, volatile fraction and organoleptic characteristics of a white-brined goat milk cheese. Small Rumin Res 145:1–6
42. Mallatou H, Pappas CP, Voutsinas LP (1994) Manufacture of feta cheese from sheep's milk, goat's milk or mixtures of these milks. Int Dairy J 4(7):641–664
43. Zoidou E, Plakas N, Giannopoulou D, Kotoula M, Moatsou G (2015) Effect of supplementation of brine with calcium on the feta cheese ripening. Int J Dairy Technol 68(3):420–426
44. Farahnaky A, Mousavi SH, Nasiri M (2013) Role of salt in Iranian ultrafiltered feta cheese: some textural and physicochemical changes during ripening. Int J Dairy Technol 66(3):359–365
45. Pappa EC, Kandarakis I, Zerfiridis GK, Anifantakis EM, Sotirakoglou K (2006) Influence of starter cultures on the proteolysis of teleme cheese made from different types of milk. Lait 86:273–290
46. Azambuja NC, Moreno I, Gallina DA, Spadoti LM, Motta LMP, Pacheco MTB, de Queiro ALM, Antunes AEC (2017) Effect of adjunct culture Lactobacillus helveticus (B02) on the composition, proteolysis, free amino acids release and sensory characteristics of Prato cheese. Food Nutr Sci 8:512–525
47. Moatsou G, Massouras T, Kandarakis I, Anifantakis E (2002) Evolution of proteolysis during the ripening of traditional feta cheese. Lait 82:601–611
48. Volikakis P, Biliaderis CG, Vamvakas C, Zerfiridis GK (2004) Effects of a commercial oat-β-glucan concentrate on the chemical, physic-chemical and sensory attributes of a low-fat white-brined cheese product. Food Res Int 37:83–94
49. Tungjaroenchai W, Drake MA, White CH (2001) Influence of adjunct cultures on ripening of reduced fat edam cheeses. J Dairy Sci 84(10):2117–2124
50. Molina E, Ramos M, Alonso L, López-Fandiño R (1999) Contribution of low molecular weight water soluble compounds to the taste of cheeses made of cows', ewes' and goats' milk. Int Dairy J 9(9):613–621
51. Nega A, Moatsou G (2012) Proteolysis and related enzymatic activities in ten Greek cheese varieties. Dairy Sci Technol 92(1):57–73
52. Calvo MV, Castillo I, Díaz-Barcos V, Requena T, Fontecha J (2007) Effect of hygienized rennet paste and a defined strain starter on proteolysis, texture and sensory properties of semi-hard goat cheese. Food Chem 102(3):917–924
53. Kondyli E, Katsiari MC, Masouras T, Voutsinas LP (2002) Free fatty acids and volatile compounds of low-fat feta-type cheese made with a commercial adjunct culture. Food Chem 79(2):199–205
54. Mallatou H, Pappa E, Massouras T (2003) Changes in free fatty acids during ripening of Teleme cheese made with ewes', goats', cows' or a mixture of ewes' and goats' milk. Int Dairy J 13(2):211–219
55. Atasoy AF, Turkoglu H (2008) Changes of composition and free fatty acid contents of Urfa cheeses (a white-brined Turkish cheese) during ripening: effects of heat treatments and starter cultures. Food Chem 110(3):598–604
56. Atasoy AF, Turkoglu H (2009) Lipolysis in Urfa cheese produced from raw and pasteurized goats' and cows' milk with mesophilic or thermophilic cultures during ripening. Food Chem 115(1):71–78
57. Laskaridis K, Serafeimidou A, Zlatanos S, Gylou E, Kontorepanidou E, Sagredos A (2013) Changes in fatty acid profile of feta cheese including conjugated linoleic acid. J Sci Food Agric 93(9):2130–2136
58. Atasoy AF (2015) Effect of partial substitution of caprine milk for ovine milk, dry salting and cured scalding on lipolysis in Urfa cheeses. Int J Dairy Technol 68(3):381–388
59. Poveda JM, Cabezas L (2006) Free fatty acid composition of regionally-produced Spanish goat cheese and relationship with sensory characteristics. Food Chem 95(2):307–311
60. Serhan M, Linder M, Hosri C, Fanni J (2010) Changes in proteolysis and volatile fraction during ripening of Darfiyed, a Lebanese artisanal raw goat's milk cheese. Small Rumin Res 90(1–3):75–82
61. Tabet E, Mangia NP, Mouannes E, Hassoun G, Helal Z, Deiana P (2016) Characterization of goat milk from Lebanese Baladi breed and his suitability for setting up a ripened cheese using a selected starter culture. Small Rumin Res 140:13–17
62. Marilley L, Casey MG (2004) Flavours of cheese products: metaboic pathways, analytical tools and identification of producing strains. Review article Int J Food Microbiol 90:139–159
63. Angelopoulou A, Alexandraki V, Georgalaki M, Anastasiou R, Manopoulou E, Tsakalidou E, Papadimitriou K (2017) Production of probiotic Feta cheese using Propionibacterium freudenreichii subsp. Shermanii as adjunct Int Dairy J 66:135–139
64. Bontinis ThG, Mallatou H, Pappa EC, Massouras Th, Alichanidis E (2012) Study of proteolysis, lipolysis and volatile profile of a traditional Greek goat cheese (Xinotyri) during ripening. Small Rumin Res 105:193–201
65. Lucas A, Coulon JB, Agabriel C, Chilliard Y, Rock E (2008) Relationships between the conditions of goat's milk production and the contents of some components of nutritional interest in Rocamadour cheese. Small Rumin Res 74(1–3):91–106
66. Barlowska J, Pastuszka R, Rysiak A, Król J, Brodziak A, Kedzierska - Matysek M, Wolanciuk, Litwinczuk Z, (2018) Physicochemical and sensory properties of goat cheeses and their fatty acid profile in relation to the geographic region of production. Int J Dairy Technol 71(3):699–708
67. McCarthy CM, Kelly PM, Wilkinson MG, Guinee T (2017) Effect of fat and salt reduction on the changes in the concentration of free amino acids and free fatty acids in Cheddar-style cheeses during maturation. J Food Comp Anal 59:132–140
68. Guler Z, Uraz T (2004) Relationships between proteolytic and lipolytic activity and sensory properties (taste–odour) of traditional Turkish white cheese. Int J Dairy Techn 57(4):237–242
69. Ozer B, Kirmaci HA, Hayaloglu AA, Akcelik M, Akkoc N (2011) The effects of incorporating wild-type strains of Lactococcus lactis into Turkish white-brined cheese (Beyaz peynir) on the fatty acid and volatile content. Int J Dairy Techn 64(4):494–501
70. Guizani N, Al-Attabi Z, Kasapis S, Gaafar OM (2006) Ripening profile of semi-hard standard goat cheese made from pasteurized milk. Int J Food Properties 9(3):523–532
71. Sheehan JJ, Patel AD, Drake MA, McSweeney PLH (2009) Effect of partial or total substitution of bovine for caprine milk on the compositional, volatile, non-volatile and sensory characteristics of semi-hard cheeses. Int Dairy J 19(9):498–509
72. Sipahioglu O, Alvarez VB, Solano – Lopez C, (1999) Structure, physico-chemical and sensory properties of feta cheese made with tapioca starch and lecithin as fat mimetics. Int Dairy J 9:783–789
73. Półtorak A, Wyrwisz J, Moczkowska M, Marcinkowska-Lesiak M, Stelmasiak A, Ulanicka U, Zalewska M, Wierzbicka A, Sun D-W (2015) Correlation between instrumental texture and colour quality attributes with sensory analysis of selected cheeses as affected by fat contents. Int J Food Sci Technol 50(4):999–1008
74. Pappa EC, Kandarakis I, Mallatou H (2007) Effect of different types of milks and cultures on the rheological characteristics of Teleme cheese. J Food Eng 79(1):143–149

75. Kumar S, Kanawjia SK, Kumar S (2015) Incorporation of Lactobacillus adjuncts culture to improve the quality of Feta-type cheese made by using buffalo milk. J Food Sci Techn 52(8):5021–5029
76. Karami M, Ehsani MR, Mousavi SM, Rezaei K, Safari M (2009) Changes in the rheological properties of Iranian UF-Feta cheese during ripening. Food Chem 112(3):539–544
77. Tsigkros D, Folland E, Moate R, Brennan CS (2003) Feta cheese texture: the effect of caprine and ovine milk concentration. Int Dairy J 56(4):233–236
78. Koca N, Balasubramaniam VM, Harper WJ (2011) High-pressure effects on the microstructure, texture and color of white-brined cheese. J Food Sci 76(5):399–404