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Characterisation of Urda whey cheese: Evolution of main biochemical and microbiological parameters during ripening and vacuum packaged cold storage

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ABSTRACT

In this study, the main biochemical and microbiological characteristics of Urda, a traditional Greek whey cheese, were determined during ripening at 19 ± 2 °C for 25 days followed by vacuum packaging and storage at 5 °C until day 360. Few differences in pH, water activity, acid degree value, moisture, salt, protein and fat contents were observed between Urda cheeses produced from sheep or goat milk whey at all sampling days (1, 25, 90, 180 and 360). Cheese microbiota was dominated by mesophilic lactic acid bacteria, but high numbers of enterococci, aerobic gram-negative bacteria and Enterobacteriaceae were also present. Increases in all the soluble nitrogen fractions of cheeses occurred, primarily during cold storage. Ketones and terpenes were the most abundant volatile compounds in fresh (1-day) sheep and goat cheeses, respectively, whereas free fatty acids were the most abundant compounds in mature (180-day) cheeses, followed by ketones.

1. Introduction

Traditional cheese production has strong, positive, social and economic impacts in terms of maintaining local employment and retaining farmers and their families in rural areas. This is particularly important for Greece, which has many mountainous or dry island areas (Bontinis, Mallatou, Alichanidis, Kakouri, & Samelis, 2008; Kondyli, Svarnas, Samelis, & Katsiari, 2012a).

Urda is a traditional Greek cheese produced from sheep or goat milk whey, but it has not been studied or characterised. It is mainly manufactured as a farmhouse cheese, in high altitude (1500 m) mountainous areas of the region of Konitsa, Epirus, during summer. Urda is consumed either as fresh or mature cheese. The cheese can be air-ripened in cool farm places, such as cellars, storehouses, shelters, for 20–30 days and then air-stored in a refrigerator or cold places for up to one year. The mature cheese has a rich aroma and taste and is of high consumer acceptance in local markets. The objective of this study was to investigate the main biochemical and microbiological characteristics of Urda cheese.

2. Materials and methods

2.1. Cheese manufacture and sampling

All Urda cheeses of this study were made in a farmhouse of the village of Aetomilitsa, Konitsa, following the same traditional manufacturing protocol and using simple processing equipment. The whey used was obtained after manufacture of hard cheese from raw sheep or goat milk with a procedure that included “beating” the coagulum to enrich the whey with fat.

The drained sheep whey (55 kg) or goat whey (65 kg) was then filtered and heated gradually under continuous stirring to 55 °C. At this point sheep milk (12 kg) or goat milk (22 kg) was added to the whey, and heating under stirring was continued. At 70 °C salt (1%, w/v) was added, and heating was continued up to 80 °C under gentle stirring. When small curd particles of the coagulated whey proteins appeared, stirring was stopped. The curd was cooked at 80 °C for 20 min and then transferred to pierced stainless steel moulds containing a cheese cloth, which was tied up to form the cheeses. Ten cheese moulds were formed from every sheep or goat milk whey batch. Then the moulds were removed and the cheeses were left to drain at ambient temperature overnight. On the next day the cheeses (approximately 800 g each) were washed with

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brine and then dry salted. After salting, cheeses were ripened on wooden shelves in a naturally ventilated room at 19 ± 2 °C and 70–75% relative humidity (RH) for 25 days. Then the mature cheeses were vacuum packaged in polyethylene bags (AlexPak, Ioannina, Greece) to prevent mould growth and excessive moisture loss, and stored at 5 °C until day 360 after manufacture. Samples were taken for analyses at 1, 25, 90, 180 and 360 days after cheese manufacture. Three individual Urda cheese making trials from sheep whey and another three from goat whey manufactured on three consecutive summer days were studied by analysing two samples per trial at each sampling day.

2.2. Physicochemical analysis

Cheese samples were analysed for moisture, salt, fat and protein content, pH, water activity (a_w) and acid degree value (ADV) as described by Bontinis et al. (2008). Total nitrogen (TN), soluble nitrogen (SN) fractions, i.e., water-soluble nitrogen (WSN), nitrogen soluble in 12% trichloroacetic acid (TCA-N) or in 5% phosphotungstic acid (PTA-N), free fatty acids (FFAs) and other volatile compounds were determined as described by Kondyli, Pappa, and Vlachou (2012b). Cheese mineral elements were determined according to Pappas et al. (2011).

2.3. Microbiological analysis

Cheese samples (25 g) were transferred aseptically to stomacher bags, then 225 mL of 0.1% buffered peptone water (Merck, Darmstadt, Germany) were added and the mixture was homogenised in a stomacher (Lab Blender, Seward, London, UK) for 60 s at room temperature. Homogenates were analysed for total mesophilic bacteria, mesophilic and thermophilic lactic acid bacteria (LAB), mesophilic and thermophilic dairy cocci, enterococci, aerobic gram-negative bacteria, total Enterobacteriaceae, total staphylococci and other catalase-positive bacteria and yeasts, according to the procedures reported by Bontinis et al. (2008).

2.4. Statistical analysis

Multifactorial analysis of variance was used to compare the values of each parameter of Urda cheese made using sheep or goat whey at each sampling day. Moreover, a comparison of the values of each parameter of Urda cheese made with one type of whey at different ages was made. The software Statgraphics Plus for Windows (version 5.2, Manugistics, Inc., Rockville, MD, USA) was used. Means and standard error values were calculated and when F values were significant at the $P < 0.05$ level, mean differences were compared by the least significance difference (LSD) procedure.

3. Results and discussion

Table 1 summarises the changes in the main physicochemical parameters of Urda cheese during ripening and cold storage. A major decrease in the cheese pH accompanied by a significant loss of moisture and a consequent decrease in a_w values ($P < 0.05$) were observed in all cheeses from days 1–25. The moisture content of mature cheeses did not differ significantly between sheep cheese and goat cheese. Similar moisture contents were reported by Govaris, Koidis, and Papatheodorou (2001) for Manouri cheese whereas Kaminarides, Nestoratos, and Massouras (2013) found higher moisture contents in fresh (1-day) sheep whey cheeses than those found in this study. As expected, the salt, fat and protein contents of the cheeses increased during ripening ($P < 0.05$), and remained almost constant ($P > 0.05$) during cold storage. Salt, fat and protein contents of sheep and goat cheeses did not differ significantly at any of the sampling days. The mature cheeses contained 3.5–4% salt, while their protein contents were similar to those reported for other whey cheeses (Kaminarides et al., 2013).

ADV value was used as an index of lipolysis. Regardless of the type of whey used, ADV increased during ripening ($P < 0.05$). ADV of sheep and goat cheeses did not differ significantly at any of the sampling days (Table 1). ADVs of Urda were generally higher than those reported for other whey cheese varieties (Kavas & Kavas, 2011; Lioliou, Litopoulou-Tzanetaki, Tzanetakis, & Robinson, 2001).

Microbial counts (Table 2) showed that fresh (day 1) Urda cheeses contained a diverse adventitious microbiota, which mainly included different types of LAB, enterococci, and Enterobacteriaceae. Mesophilic LAB was the most prevalent group in fresh cheeses with their initial contamination ranging at a level of 5 log cfu g^{-1} and 4 log cfu g^{-1} in sheep and goat cheeses, respectively (Table 2). Enterobacteriaceae (oxidase-negative colonies) also grew at similar levels on the *Pseudomonas* agar plates used to enumerate aerobic Gram-negative bacteria (oxidase-positive, pigment colonies) that were also present in all fresh (day 1) cheeses at an initial contamination level of 5.0 log cfu g^{-1} (data not shown). Staphylococci and other types of catalase-positive bacteria were present at an initial contamination level of 4 log cfu g^{-1}, while yeasts were below 2 log cfu g^{-1} in all cheeses on day 1 (data not shown).

Table 1
Changes of physicochemical characteristics of Urda cheese made from sheep (S) or goat (G) milk whey during ripening and storage.[a]

Parameter	Cheese type	Cheese age (days)				
		1	25	90	180	360
Moisture (%)	S	56.97 ± 2.27^{a}	30.54 ± 1.09^{b}	30.76 ± 1.27^{b}	31.05 ± 1.08^{b}	29.82 ± 1.06^{b}
	G	54.13 ± 0.41^{a}	27.40 ± 0.89^{b}	27.52 ± 0.70^{b}	28.27 ± 0.64^{b}	28.04 ± 0.52^{b}
NaCl (%)	S	0.60 ± 0.06^{a}	3.83 ± 0.18^{b}	3.64 ± 0.19^{b}	3.88 ± 0.18^{b}	3.53 ± 0.14^{b}
	G	0.51 ± 0.01^{a}	3.50 ± 0.17^{bc}	3.35 ± 0.13^{bc}	3.77 ± 0.21^{c}	3.07 ± 0.19^{b}
Fat (%)	S	28.8 ± 0.0^{a}	42.8 ± 1.3^{b}	43.5 ± 1.5^{b}	43.0 ± 1.0^{b}	44.0 ± 2.0^{b}
	G	29.7 ± 0.8^{a}	46.0 ± 1.5^{b}	45.7 ± 1.3^{b}	44.7 ± 1.9^{b}	46.9 ± 2.6^{b}
Proteins (%)	S	14.57 ± 0.81^{a}	20.13 ± 0.67^{b}	21.92 ± 0.26^{b*}	19.94 ± 0.67^{b}	20.12 ± 0.02^{b}
	G	12.63 ± 0.14^{a}	18.77 ± 0.11^{b}	18.38 ± 0.09^{b*}	18.36 ± 0.36^{b}	18.22 ± 0.85^{b}
pH	S	6.39 ± 0.01^{a}	5.28 ± 0.08^{b}	5.04 ± 0.04^{bc}	4.99 ± 0.13^{c}	5.27 ± 0.07^{b}
	G	6.41 ± 0.01^{a}	5.56 ± 0.13^{b}	5.46 ± 0.16^{b}	5.23 ± 0.08^{b}	5.27 ± 0.13^{b}
a_w	S	0.948 ± 0.001^{a*}	0.936 ± 0.004^{b}	0.929 ± 0.005^{b}	0.900 ± 0.005^{c}	0.899 ± 0.001^{c}
	G	0.965 ± 0.004^{a*}	0.919 ± 0.008^{bc}	0.928 ± 0.004^{b}	0.909 ± 0.005^{cd}	0.905 ± 0.001^{d}
ADV	S	0.25 ± 0.03^{a}	0.70 ± 0.09^{b}	0.71 ± 0.1^{bc}	1.08 ± 0.03^{bc}	1.25 ± 0.05^{c}
	G	0.28 ± 0.02^{a}	0.54 ± 0.02b^{b}	0.52 ± 0.07^{b}	0.90 ± 0.05^{c}	0.88 ± 0.06^{c}

[a] Abbreviations are: a_w, water activity; ADV, acid degree value (meq KOH 100 g^{-1} fat). Values are means of three cheese-making trials ± standard error; means for each parameter in the same column and sampling day with an asterisk are significantly different ($P < 0.05$); means in the same row with different superscript lower case letter are significantly different ($P < 0.05$).