UFT Academic Publishing House, Plovdiv

Food Science and Applied Biotechnology
e-ISSN: 2603-3380

Journal home page: www.ijfsab.com
https://doi.org/10.30721/fsab2023.v6.i1

OPEN ACCESS

Research Article

Comparative study of quality characteristics of Kashkaval cheese from fresh and chilled curd

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Abstract
The influence of chilled curd on Kashkaval cheese microbiological, functional, textural and sensorial characteristics was studied. It was found that salting the curd in a hot solution influenced to a greater degree the microflora reduction from the starter culture in Kashkaval cheese obtained from fresh curd. In terms of species composition, *Streptococcus* ssp. had a higher survival rate compared to *Lactobacillus* ssp. During maturation, this trend changed and the number of *Lactobacillus* ssp. increased, while that of Streptococcus ssp. remained constant and even slightly decreased in both studied samples. Melting and textural characteristics of the two studied cheese samples did not differ significantly at the end of the maturation process. The overall scores of the sensory profile were higher in the cheese obtained from fresh curd but no statistical differences ($p>0.05$) between separate sensory indices were established. The obtained results indicated that "Cagliata" can be successfully used as an alternative raw material for fresh curd in the production of Kashkaval cheese.

Keywords
chilled curd, Kashkaval cheese, cagliata cheese, pasta filata cheese, texture

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Article history:
Received 14 November 2022
Reviewed 15 December 2022
Accepted 11 February 2023
Available on-line 17 March 2023

https://doi.org/10.30721/fsab2023.v6.i1.240

Introduction

Modern science is trying to find innovative and effective solutions to problems arising from practice. The steadily increasing cow's milk production in some countries requires new and advanced approaches for its utilization. The production of chilled curd, the so-called "Cagliata" cheese, and its application in the production of different types of cheese is such a solution.

Kashkaval cheese is a hard-ripened cheese, a variety of pasta-filata cheeses, aged for 60 days before consumption (Mahon and Oberg 2017). Possibilities for its production from frozen curd, as well as its quality indicators after the ripening process, were previously reported in the literature (Simov and Ivanov 2005; Berisha et al. 2018).

The production of cheese from stored curd in a chilled or frozen state implies changes to some of the cheese properties. It was found that pasta-filata sensory profile was affected by different factors such as the application of different starter cultures (Öründü and Tarakçı 2021), the impact of packaging (Todaro et al. 2018), the aging period (Jimenez-Maroto et al. 2016), storage at different temperatures (Ivanov and Markova 2020), etc.

Various research teams reported that cheese microflora in this group was influenced by the raw material (Guidone et al. 2015; Pappa et al. 2019), the selected starter bacteria (Reale et al. 2019), and the parameters of hot salting solution (Melilli et al. 2004), etc. The influence of chilled curd on chemical characteristics was evaluated by Ivanova et al. (2021). Based on our knowledge, no data were found in the scientific literature on the sensory, microbiological and textural quality characteristics of Kashkaval made from chilled curd. Therefore, the aim of the present research was to study the influence of chilled curd on quality characteristics of Kashkaval cheese produced from "Cagliata" as a raw material.

Materials and Methods

Materials. Chilled and fresh curd, obtained from fresh cow's milk, with a known chemical composition were used in order to prepare Kashkaval cheese (Ivanova et al. 2021). In brief, dry matter, milk fat, total protein, ash, milk fat and pH of the two types of curd were determined. The microbiological initial status of chilled and fresh curd is presented in Table 1.

Table 1. Microbiological analysis of fresh and chilled curd

Component	Chilled curd	Fresh curd
Lactobacillus ssp., log cfu/g	4.67±0.30[a]	6.71±0.50[b]
Streptococcus ssp., log cfu/g	4.59±0.40[a]	6.69±0.30[b]
Coliforms, log cfu/g	-	-
Moulds and yeasts, cfu/g	-	-

[a,b] Means with different letters within a row are significantly different ($p<0.05$)

Cheese preparation. Procedures for cheese preparation were described in full in the companion paper (Ivanova et al. 2021). In brief, two types of cheeses were prepared – one was obtained from fresh cow milk curd and the second was prepared from chilled curd (1°C, 2 months) according to the technology previously described in full (Ivanova et al. 2021).

Microbiological analysis. The preparation of cheese samples for microbiological examination was conducted in accordance with BNS EN ISO 6887-5:2020 (2020) procedure.

The total number of *Lactobacillus* ssp. was determined by BNS EN ISO 9232:2005 (2005).

The number of *Streptococcus* ssp. was evaluated in correspondence with BNS EN ISO 7889:2005 (2005) procedure. They were calculated by selective synthetic nutrient media of MRS (Merck) and M17 (Merck) respectively.

Moulds and yeasts were detected by the BNS EN ISO 6611:2006 (2006) standard by deep sowing in Yeast Extract Glucose Chloramphenicol Agar (Merck).

Coliform bacteria and *E. coli* were determined by chromogenic agar for coliforms Chromocult (Merck) according to ISO 4832:2006 (2006) method.

Melting characteristics. The meltability of the cheese samples was evaluated by the modified Schreiber test as described by Sharma et al. (2015). 5 g cylindrical cheese probes with a diameter of 47 mm and thickness of 2 mm were cut from a block of cheese using a bore and placed in a petri dish. The petri dish was covered, placed at 4°C for 10 min for temperature equilibration, then placed for 5 min in a forced-air convection oven at 232°C. After removing from the oven, molten cheese samples were cooled on a flat surface for 30 min. A Nikon D5600 digital SLR camera with a 30 mm focal length and a shutter speed of 1/160 s was used to capture digital images of the cheese melt area. The extent of melt spread expressed as melt score in % was calculated using image analysis (ImageJ). For each cheese sample, the meltability test was conducted in quadruplicate.

Textural analysis. Cheese hardness was measured with texture analyzer Stevens-LFRA (Brookfield Engineering Lab. Ing, Massachusetts, USA) based on the modified method AACC 74-09 (1988). The compressing cylinder with a diameter of 10mm penetrated to a depth of 3 mm with a crosshead speed of 1mm.s^{-1}. The maximal penetration force was measured in g. The relative elasticity was further determined by measuring the remaining force after 32 s retention time during compression at a maximum penetration depth of 3 mm. The relative elasticity was defined as a ratio of the remaining compression force at 32 s retention time compared to the maximum measured compression force. It has been established that the change in the pH value of the paddlefish fillet during its 7d storage at 0 - 4°C practically does not depend on the presence of an alginate coating and a surface treatment with DDRPE or L-ascorbic acid solutions.

Colour characteristics. The colour characteristics of the tested samples were evaluated by laboratory colorimeter PCE-CSM 2 (Germany) using CIELab colour system. The total colour differences dE among the samples was calculated according to:

$$\boldsymbol{dE = \sqrt{(L* - L_0)^2 + (a* - a_0)^2 + (b* - b_0)^2}} \quad (1)$$

where: L*, *a**, *b** are colour characteristics of the sample at the end of the ripening period, and L_0, a_0, b_0 colour characteristics of the sample at the beginning of the ripening period.

Sensory analysis. The sensory analysis of Kashkaval cheeses was performed at the beginning and at the end of the repining period (60th d) in accordance with the Bulgarian National Standard BNS 15612:1983 (1983) by 15 untrained panellists in order to evaluate consumer acceptance. The evaluation criteria were the following: taste and aroma - 45 points, consistency - 20 points, structure and cut surface - 15 points, appearance - 5 points, colour - 5 points and packaging – 10 points (maximum overall score - 100 points).

Statistical analysis. Microsoft Excel 2010 (ANOVA) was used for computer processing of the obtained data. Multiple comparisons were made according to the LSD (Lowest Standard Deviation) procedure. Results are expressed as the mean ± SD (standard deviation) with four replications (n=4). They were considered as significantly different when $p<0.05$.

Results and Discussion

Microbiological analysis. Different lactic acid microorganisms have different sensitivities to salt and temperature treatment. The results obtained from the microbiological analysis are presented in Table 2. Salting the curd in a hot water solution (at 72°C and 14% salt content) kills much of its microflora. Our research demonstrated that there was a significant reduction ($p<0.05$) of *Lactobacillus ssp*. in Kashkaval cheese produced from chilled curd (30%) and even bigger shown in Kashkaval cheese produced from fresh curd (56%). This can be explained by the fact that the chilled curd already reduced partially its microflora during chilled storage and fresh curd was still rich in microflora because it was analyzed immediately after cheddaring. The number of *Streptococcus* ssp. was reduced by 14 % in the case of chilled curd and by 26 % when cheese was obtained from fresh curd. In this case, the greater reduction of microflora in

the cheese made from fresh cheese was also confirmed. It was found that salt concentration beyond 6.5% in the water phase of the curd stopped the development of the starter culture (Kozhev 2006).

Our research established that *Streptococcus* ssp. had a higher survival rate than *Lactobacillus* ssp., a tendency confirmed also by other authors (Gruev 1995). A significant increase ($p<0.05$) in the number of viable *Lactobacillus* ssp. was found in both Kashkaval cheese samples during the ripening period.

The same tendency was reported by Simov and Ivanov (2005). The number of viable cells of *Steptococcus* ssp. increased slightly (Kashkaval obtained from fresh curd) or remained constant (Kashkaval obtained from chilled curd). A similar tendency was observed by Pappa et al. (2019) for Kashkaval cheese produced from pasteurized ewe's milk. No coliforms, yeasts and moulds were detected in both Kashkaval cheeses. This was important to demonstrate the possible safe and quality production of Kashkaval cheese when chilled curd was used instead of fresh curd as mentioned by Losito et al. (2014) for the pasta-filata type cheeses.

Table 2. Microbiological analysis of Kashkaval from chilled and fresh curd

Component	**Kashkaval from chilled curd**		**Kashkaval from fresh curd**	
	Beginning of ripening	End of ripening (60th d)	Beginning of ripening	End of ripening (60th d)
Lactobacillus ssp., log cfu.g^{-1}	3.29 ± 0.30^{a}	4.12 ± 0.30^{b}	2.96 ± 0.22^{a}	4.22 ± 0.25^{b}
Streptococcus ssp., log cfu.g^{-1}	3.97 ± 0.20^{a}	3.70 ± 0.20^{a}	4.94 ± 0.12^{b}	3.77 ± 0.24^{a}
Coliforms, log cfu.g^{-1}	-	-	-	-
Moulds and yeasts, cfu.g^{-1}	< 10	< 10	-	-

$^{a, b}$ *Means with different letters within a row are significantly different ($p<0.05$)*

Melting properties. When cheese is subjected to baking the bonds holding together the casein proteins break and the cheese melts down into a thick fluid (cheese melt). Many factors affect melting ability but one of the most important is moisture content. High-moisture cheeses, like pasta-filata flow more easily than dry hard cheeses. In moist cheese, the proteins are loosely packed with water pools interspersed between them, so they readily liquefy. Fig. 1 represents the initial cheese sample and cheese melt after baking following the Schreiber melt test procedure.

Table 3 represents the Shreiber melt score for Kashkaval cheese manufactured from chilled and fresh curd at the beginning and at the end of the ripening period. The results indicated an increase in the melt score for the cheese from fresh curd with ripening time, and on the 60th d, the melt score of the chilled and fresh curd converged and approached similar values. At the end of the ripening period (the 60th d) the relative elasticity of the Kashkaval manufactured from chilled curd decreased and reached values similar to the samples manufactured with fresh curd. However, at the end of the ripening period hardness of the Kashkaval from chilled curd was about 25% higher compared to Kashkaval from fresh curd.

Textural properties (hardness and relative elasticity) Table 4 represents the textural properties of Kashkaval manufactured from chilled and fresh curd during the ripening period of 60 d. Both hardness and relative elasticity of samples manufactured from fresh curd decreased over time, similarly to the results reported previously in the

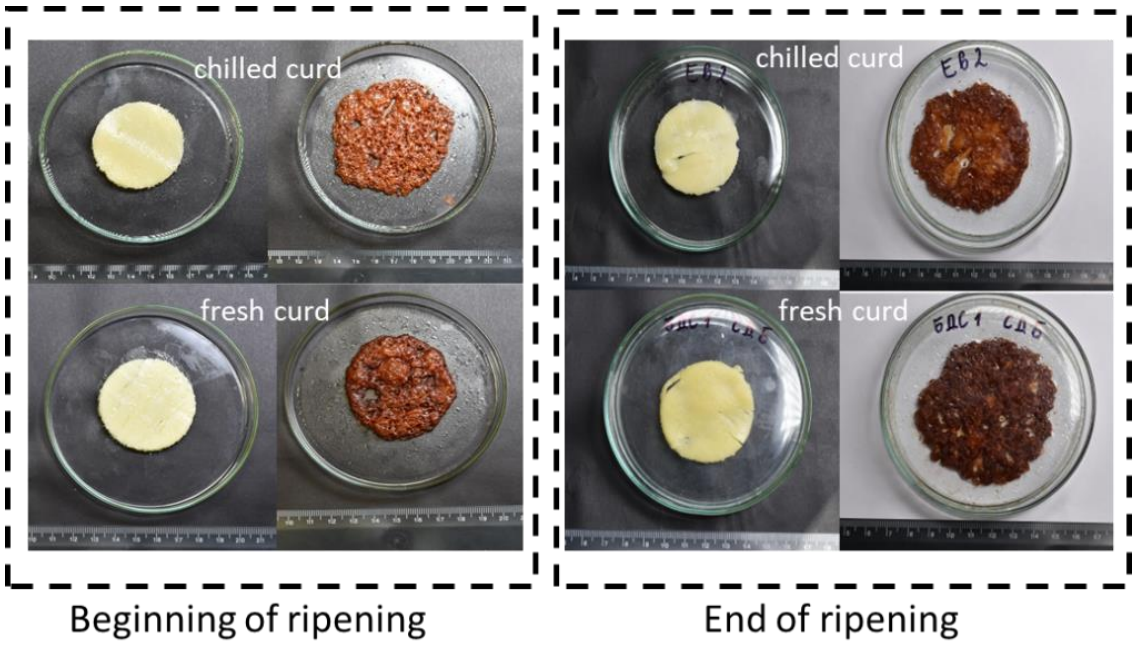


Figure 1. Schreiber melt test – cheese samples before (yellowish) and after (brownish) melting

literature (Fuentes et al. 2015). The overall score at the beginning of the ripening process was 86.2 points for the cheese manufactured from chilled curd and 90.0 for the cheese produced from fresh curd. Similar results were found by Fuentes et al. (2015) who explained the gradual loss of quality with some structural characteristics such as a decrease in hardness and appearance of off-flavors in non-aged pasta-filata cheese. The established tendency in our research continued after 60 d when the second sensory evaluation was done.

These results are comparable with those obtained by Sandoval-Copado et al. (2016) who found a strong correlation between specific sensory data and chemical characteristics of cheese from the pasta-filata family. At this stage, the cheese from "Cagliata" obtained 91.80 points and the classic cheese 94.33 respectively. The higher overall scores were due to the ripening process which attributed to the specific sensory profile of the cheese. Cooking salt improved the consistency of Kashkaval cheese, as it caused swelling and peptization of paracasein (Kozhev 2006).

Table 3. Melt score of Kashkaval from chilled and fresh curd

Parameter	**Kashkaval from chilled curd**		**Kashkaval from fresh curd**	
	Beginning of ripening	End of ripening (60th d)	Beginning of ripening	End of ripening (60th d)
Melt score, %	183.6 ± 11.7^{a}	178 ± 2.04^{a}	129.5 ± 5.93^{b}	193.6 ± 23.21^{ab}

$^{a, b}$ *Means with different letters within a row are significantly different ($p<0.05$)*

A comparison of the two maturation periods showed that all indices increased significantly ($p<0.05$). No statistical differences ($p>0.05$) between samples were found when the single indices of the sensory profiles were compared for the same period. This was a good indicator for consumer acceptance of both products at the end of the ripening period. At the end of the ripening period (60th d), the relative elasticity of the Kashkaval manufactured from chilled curd decreased and reached values similar to

the samples manufactured with fresh curd. However, at the end of the ripening period hardness of the Kashkaval from chilled curd was about 25% higher compared to Kashkaval from fresh curd.

Colour characteristics. Colour characteristics (CIELab and dE) of the Kashkaval samples manufactured from chilled and fresh curd are presented in Table 5.

Table 4. Texture properties of Kashkaval from chilled and fresh curd

Parameter	**Kashkaval from chilled curd**		**Kashkaval from fresh curd**	
Texture characteristics	Beginning of ripening	End of ripening (60^{th} d)	Beginning of ripening	End of ripening (60^{th} d)
Hardness (maximal compression force), g	582 ± 141.1^{a}	618 ± 76.8^{a}	942 ± 157.7^{b}	470 ± 46.2^{ab}
Relative elasticity, %	48.1 ± 4.04^{a}	47.5 ± 1.75^{a}	48.9 ± 2.53^{a}	43.8 ± 0.88^{b}

$^{a, b}$ Means with different letters within a row are significantly different ($p<0.05$)

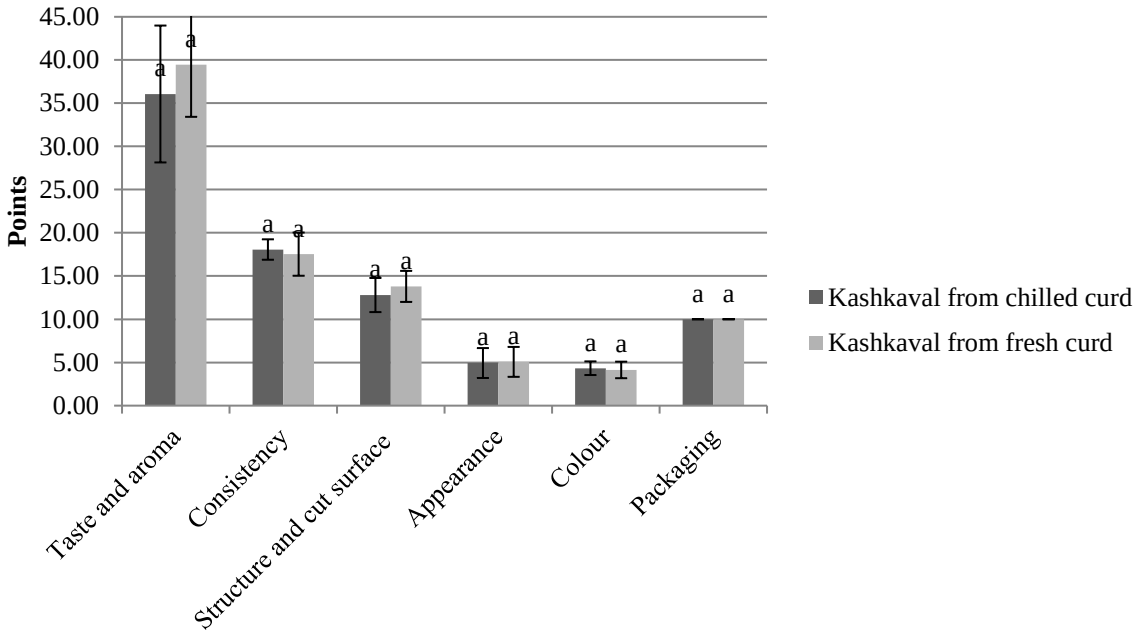
The lightness (L) of the Kashkaval sample manufactured from chilled curd was lower at the beginning of the ripening process compared to the fresh curd cheese but at the 60^{th} d both samples ended up with similar L values. According to the total colour change indicator dE, the Kashkaval from chilled curd undergoes lower colour changes during ripening compared to Kashkaval from fresh curd.

Sensory analysis. The sensory profiles of Kashkaval from chilled and fresh curd at the beginning and at the end of the ripening day of the ripening period are presented in Fig. 2.

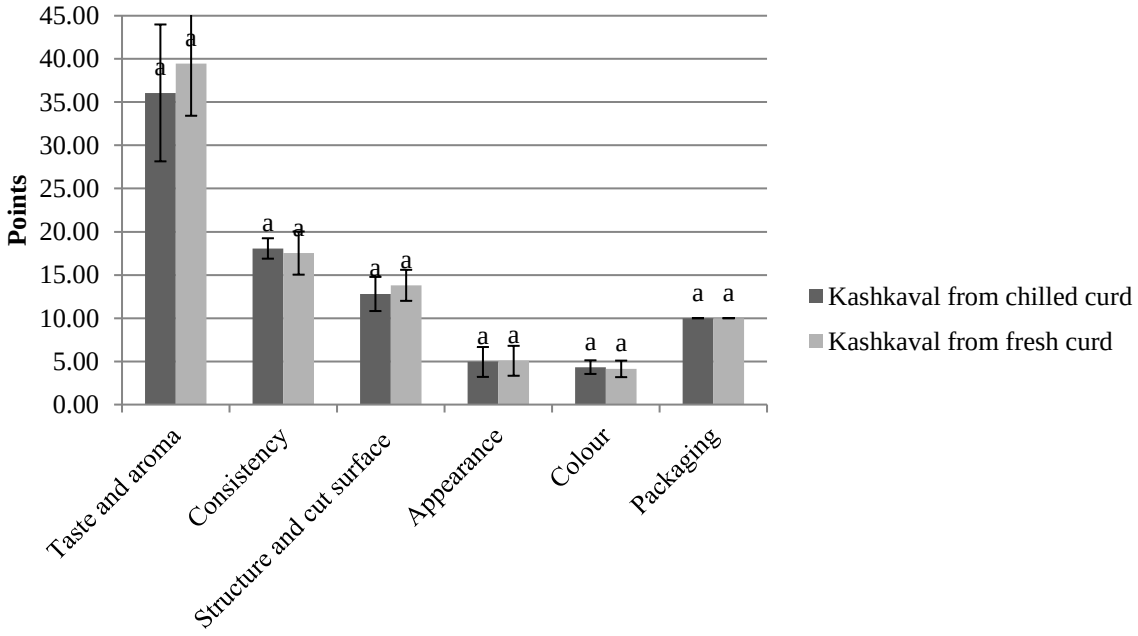
Table 5. Color characteristics of Kashkaval from chilled and fresh curd

Parameter	**Kashkaval from chilled curd**		**Kashkaval from fresh curd**	
Colour characteristics	Beginning of ripening	End of ripening (60^{th} d)	Beginning of ripening	End of ripening (60^{th} d)
Lightness (L)	70.7 ± 6.07^{a}	75.7 ± 0.37^{a}	84.09 ± 1.46^{b}	79.9 ± 0.69^{ab}
Redness (a)	0.86 ± 0.15	0.28 ± 0.21	-0.13 ± 0.07	-0.13 ± 0.02
Yellowness (b)	22.7 ± 1.43	24.4 ± 0.08	26.8 ± 0.96	31.35 ± 0.45
Chroma (C)	22.7 ± 1.41	24.4 ± 0.08	26.5 ± 1.2	31.35 ± 0.45
Hue (H)	87.5 ± 0.005	89.37 ± 0.47	90.3 ± 0.36	87.25 ± 4.41
Total colour differences dE	5.4^{a}		6.18^{b}	

$^{a, b}$ Means with different letters within a row are significantly different ($p<0.05$)



At the beginning of the ripening period (A)



At the end of the ripening (60th d) period (B)

Figure 2. Sensory profile of Kashkaval from chilled and fresh curd at the beginning (A) and in the end of the ripening (60th d) period (B)

The overall score at the beginning of the ripening process was 86.2 points for the cheese manufactured from chilled curd and 90.0 for the cheese produced from fresh curd. Similar results were found by Fuentes et al. (2015) who explained the gradual loss of quality with some structural characteristics such as a decrease in hardness and the appearance of off-flavors in non-aged pasta-filata cheese. The established tendency in our research continued after 60 d when the second sensory evaluation was done. At this stage, the cheese from "Cagliata" obtained 91.80 points and the classic cheese 94.33 respectively. The higher overall scores were due to the ripening process which attributed to the specific sensory profile of the cheese. Cooking salt improved the consistency of the cheese, incl. Kashkaval cheese, as it caused swelling and peptization of paracasein (Kozhev, 2006). A comparison of the two maturation periods showed that all indices increased significantly ($p<0.05$). These results were comparable with those obtained by Sandoval-Copado et al. (2016) who found a strong correlation between specific sensory data and chemical characteristics of cheese from the pasta-filata family. No statistical differences ($p>0.05$) between samples were found when the single indices of the sensory profiles were compared for the same period. This was a good indicator for consumer acceptance of both products at the end of the ripening period.

Conclusions

The obtained results demonstrated that chilled curd instead of fresh curd can be successfully used in the production of Kashkaval cheese. The high-quality "Cagliata" is a very good alternative to the traditional manufacturing process with no deterioration of the microbiological, functional, textural and sensory profile of the obtained cheese.

Acknowledgment

We acknowledge Liliya Kuneva and Mariana Topchieva for their technical support during this research as well as Mr. Todor Todorov, director of the company "Mlechni Producti Trakia" Ltd. - MP "Serdika Haskovo" for the provided production area for this study.

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