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Review article

The effects of soaking in salted blackcurrant wine on the properties of cheese

László Gyenge, Kinga Erdő, Csilla Albert, Éva Laslo, Rozália-Veronika Salamon *

Sapientia Hungarian University of Transylvania, Faculty of Economics, Socio-Human Sciences and Engineering, Department of Food Science, Libertății Sq. 1, 530104, Miercurea Ciuc, Romania

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ABSTRACT

The study assessed the effects of soaking hard cheeses and cheese slices in blackcurrant wine on their antioxidant, microbiological, and sensory properties. Soaking increased weekly, the phenolic content by 40.86 ± 0.3 mg GAE/100 g (gallic acid equivalent) of cheese, and their reached the maximum ethanol content of 1.29 ± 0.08 g/100 g. Soaking reduced the amount of foodborne pathogenic bacteria, enhancing microbiological safety. The lactic acid bacteria count significantly decreased during soaking time. The sensory evaluation shows that 45 % of tasters preferred the soaked cheese. Positive correlations were observed between taste and color (in whole-soaked cheese), while negative correlations were revealed among color, smell, and texture (in soaked quarter cheese). The results suggest that blackcurrant wine, containing a wealth of bioactive constituents, has the capacity to augment the favorable attributes of cheese and presents prospects for developing a novel gastronomic offering.

1. Introduction

Cheese, a highly consumed dairy product, reflecting strong demand [1], offers diverse tastes and valuable compound, including essential amino acids, fatty acids, vitamins, and minerals. Its health benefits include potential cancer prevention [2] and cholesterol reduction due to health-promoting microorganisms [3,4]. Cheese's microflora composition varies with type, processing, and ripening conditions [3,5]. Despite low functional and bioactive components, there is potential for producing high-value functional foods from cheese [6,7].

Producing functional foods with high added value can command higher prices from health-conscious consumers who appreciate the beneficial effects of such products [8]. Functional components, directly added (e.g., *Bifidobacteria*, *Lactobacteria*) or transmitted through manufacturing can improve their beneficial properties [3,9]. Efforts are also underway to create innovative products using unconventional techniques [7,10]. In this scenario, an established functional food, wine, is employed to enrich the functional qualities of cheese through soaking [11–14]. The cheese, named "Imbriago," is predominantly crafted in north-eastern Italy, traditionally incorporating grape wine or pomaces, as seen in examples like "Ubriaco di Rabosa" [15] and "Murcia al vino" [16]. Innocente et al. [17] examined how soaking traditional semi-hard cheese post-processing affects its composition, volatile compounds, proteolysis, and fatty acids. Soaking cheese in wine does not alter proteolysis or fermentation during ripening. Alcohols and short-chain ethyl esters contribute to unique fruity and wine-like aromas driven by concentration gradients between wine and cheese.

* Corresponding author.
E-mail addresses: gyengelaszlo@uni.sapientia.ro (L. Gyenge), erdokinga@yahoo.com (K. Erdő), albertcsilla@uni.sapientia.ro (C. Albert), lasloeva@uni.sapientia.ro (É. Laslo), salamonrozalia@uni.sapientia.ro (R.-V. Salamon).

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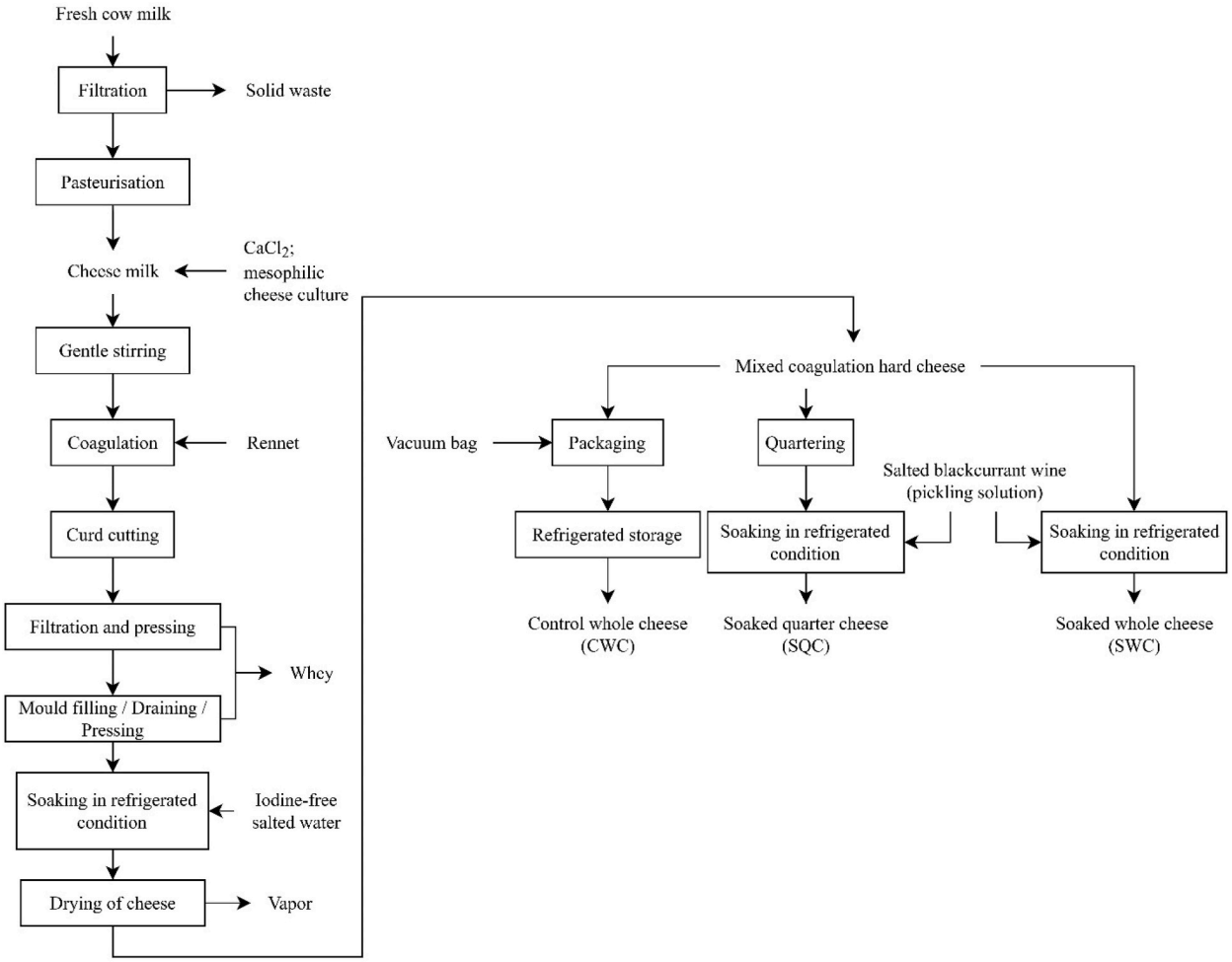


Fig. 1. Fresh milk processing into soaked and control cheese.

The blackcurrant fermented beverage produced from blackcurrant fruit (*Ribes nigrum* L.) juice [18] is rich in bioactive polyphenols (520–1985 mg GAE/L) and flavonoids (10.4–35.5 mg GAE/L), including myricetin (7.3–22.6 mg/L) and quercetin (3.1–11.9 mg/L) [19,20]. With considerable antioxidant potential, blackcurrant wine holds strong marketing appeal as a functional food, further enhanced by its mineral and ascorbic acid content [21–24].

The abundant functional components in blackcurrant wine support the hypothesis that, these elements could be absorbed in high concentrations during the cheese-soaking process. This study aims to develop a new product – cheese soaked in blackcurrant wine as a functional food – non-existent in current production. Its functional properties will be validated quantitatively, and market viability will be assessed by seeking sensory testers' opinions for an organoleptic evaluation.

2. Materials and methods

2.1. Production technology of cheese soaked in blackcurrant wine

2.1.1. Cheese production process

Raw cheese was produced through collaboration with *Cooperativa Agricolă Csengő*, a dairy company in *Frumoasa, Harghita county, Romania*. The production process of the cheese is illustrated in Fig. 1. As raw material, cow milk (temperature of 7 °C, pH 6.66, fat content of 3.27 % (w/w), protein content of 3.12 % (w/w), dry matter content of 9.55 % (w/w), density of 1029.5 kg/m^3, freezing point of 0.542 °C, negative for antibiotic test) was collected from the farmers from the nearby area. Before its use, the mixture of raw milk was filtered. For pasteurizing, a plate heat exchanger (*Pietribias*, 3000 L/h) was used at 74 °C for 30 s holding time, followed by the enrichment of milk with 23 g of $CaCl_2$/100 L milk (*Solva*y Chemicals International). The intention was to produce mixed coagulation hard cheese. As a cheese culture 300 units of *Probat 222 mesophilic culture* (*Danisco Deutschland GmbH, Lactococcus lactis* subsp. *lactis* + *L. lactis* subsp. *cremoris* 90–99.8 %, *L. lactis* subsp. *lactis* biovar. *diacetylactis 0.1-5.0 %, Leuconostoc mesenteroides* subsp. *cremoris 0.1-5.0 %*) were used, followed by of gentle stirring (1.5 h at 32 °C). As rennet, *Ideal Ultra 1000* was used, with 45 min coagulation time. The

coagulant was shredded to the size of wheat seeds, the curd manipulation was made at temperature of 42 °C. The separation of coagulants from whey took place by filtration for 25 min and the pre-pressing procedure with 160 kg weight. In the beginning, the pH of the curd was 6.37, which decreased after pre-pressing to 5.91. The curd was dosed into round forms (with a diameter of 8.5 cm and 4 cm height) and pressed for 0.5 h, followed by the soaking procedure in iodine-free salted (*Salinen Austria AG*) water (10.5 % w/V, pH 5.2) for (8 h at 15 °C). Afterward, the cheese wheels were dried for three days at 12 °C in a ventilated ripening chamber. In the ripening experimental setup, the cheese was divided into three groups: whole wheel cheese with soaking in pickling solution (SWC), quarter cheese with soaking in pickling solution (SQC), and whole cheese packed and stored at 4 °C in vacuum bag – as control samples (CWC). Samples were taken weekly and frozen at −18 °C until analyses.

2.1.2. Preparation of blackcurrant wine for pickling solution and cheese soaking

The wine was purchased from a small winery. The wine was made from four different varieties of blackcurrant ("*Fertődi 1″*, "*Tiben*", "*Tisel*", and "*Ruben*"). The blackcurrant fruits were collected in June from Sânmartin, Harghita county, Romania, the GPS coordinates are 46° 15′ 58.3812″ N and 25° 56′ 21.7212″ E, and the juice was fermented for 6 months. According to the analysis, the ethanol concentration of the wine was 12.9 % (V/V), dry matter content was 11.15 % (w/w), sugar content was 80 g/L, and density was 1025.7 kg/m^3. For preparing the pickling solution, 5 % (w/w) NaCl was added to the wine.

Before the immersion of wheel cheese into the pickling solution, the mass of each wheel and quarter was measured. The soaking process of samples SWC and SQC were carried out in plastic containers filled with pickling solution, in which the cheese to pickling solution ratio was adjusted to 1:1.5. The containers were incubated at 4 °C in a refrigerator for three weeks. Organoleptic assay, chemical and microbiological changes were followed during the experiment on a weekly or two-week basis. During sampling, was ensured that the cheese-wine ratio during the investigation always remained at 1:1.5. During the experiment, the samples were prepared in triplicates.

2.2. Dry matter and fat content determination

To determine the dry matter (DM) content of blackcurrant wine and cheese samples, *AXIS ATS60* moisture analyzer balance was used. In the case of cheese, the moisture content was determined weekly.

The fat content of soaked and ripened cheeses was determined using the Van Gulik method according to ISO 3433:2008 [25].

2.3. Methodology of preparation of samples for analytical assays

The cheese samples were homogenized for 10 min by a blender with 70 % (V/V) methanol solution in a ratio of 1:2 based on a mass basis. After that, 3 g of the mixture were transferred into centrifuge tubes, and further 7 mL of pure methanol were added and mixed well with *IKA Vortex VG 3* for 1 min. For a better exploration of the samples, the centrifuge tubes were transferred in a *VWR USC TH* type ultrasonic bath for 15 min. To separate the phases, the tubes were centrifuged at 3461 g with a *Hettich EBA 20* centrifuge equipped with *E162* rotor-type. The supernatant was used in further analysis.

2.4. Antioxidant activity determination

Since we aimed to produce a cheese with higher antioxidant properties as a functional food, this was verified by different antioxidant capacity measuring methods. The following methods were selected to analyze the antioxidant properties of cheeses.

2.4.1. 2,2-Diphenyl-1-Picrylhydrazyl (DPPH) assay

The method of Cosmulescu et al. [26] was used to assess the DPPH free radicals-scavenging ability of samples from cheeses. DPPH free radicals-scavenging activity was calculated by the following formula:

$$E\% = 100 \cdot (A_0 - A_1)/A_0 \quad (1)$$

where: A_0 - the initial absorbance, and A_1 - the absorbance in the presence of the extract.

2.4.2. Ferric reducing antioxidant power (FRAP) assay

The antioxidant potential of cheese extract was also determined using a FRAP assay measuring the change in absorbance at $\lambda = 593$ nm due to the formation of a blue-colored Fe^{2+}-tripyridyl-triazine compound from colorless oxidized Fe^{3+}-form by the action of electron-donating antioxidants. This method was described in detail by Benzie & Strain [27].

Calibration was performed with ascorbic acid, and results were expressed as milligram of ascorbic acid equivalents (AAE) per 100 g of the dry weight of cheese. Triplicate measurements were performed for all samples. The obtained equation was:

$$y = 0.0874 \cdot x + 0.0438 \ (R^2 = 0.9955) \quad (2)$$

2.4.3. Determination of total polyphenol content (TPC)

The total polyphenol content was determined using gallic acid as a standard with the Folin–Ciocâlteu reagent. The measurements

were carried out as described by Ref. [28].

The results were expressed as milligrams of gallic acid equivalent (GAE) per 100 g of DM. For the preparation of the calibration curve 5-point calibration curve was made with the measurement of absorbance values at $\lambda = 740$ nm of gallic acid in a range of 6–60 μg/mL concentration. The obtained equation of the calibration trendline is presented in eq. (3):

$$y = 46.4720 \cdot x + 0.1850 \ (R^2 = 0.9889) \quad (3)$$

2.4.4. Determination of total anthocyanin content (TAC)

Total anthocyanin content was determined according to the pH differential method [29]. The results were calculated using eq. (4) [30] and (5). Results are expressed as mg cyanidin-3-glucoside/100 g dry matter.

$$A = [(A_{510\ nm} - A_{700\ nm})_{pH\ 1.0} - (A_{510\ nm} - A_{700\ nm})_{pH\ 4.5}] \quad (4)$$

$$TAC\ (mg/L) = A \cdot MW \cdot DF \cdot 1000/\varepsilon \cdot l \quad (5)$$

where: A - absorbance; MW - molecular weight (449.2 g/mol); DF - dilution factor; ε - molar absorption coefficient (29 600 L/(mol·cm)); l - optical path length (cm).

2.5. Determination of ethanol by HPLC

For the determination of the ethanol concentration of the soaked cheese samples, the sample supernatant (prepared as described in chapter 2.5) was filtered through a syringe filter with a pore diameter of 0.45 μm. *Agilent 1200 Infinity Series* HPLC device was used for chromatographic measurement, equipped with *Coragel 87H3* column. The mobile phase was a 0.008 N sulfuric acid solution, as a detector *RID* detector was used at a temperature of 50 °C. During the test, 20 μL of the sample was injected into the HPLC at a flow of 0.6 mL/min and a column temperature of 50 °C. The obtained equation of the calibration line is presented in eq. (6):

$$y = 31035.7407 \cdot x - 1999.2262 \ (R^2 = 0.9929) \quad (6)$$

2.6. Microbiology assays

During the experiment, microbial load of cheese and blackcurrant wine pickling solution at three different times (day 0, end of 1st week, end of 3rd week) of all sets of experiments (SWC, SQC, CWC) was determined by classic spread plate methods. The number of lactic acid bacteria from the cheese was determined on de Man, Rogosa, and Sharpe (MRS) media by spread plate technique. For the determination of *Escherichia coli, Staphylococcus aureus* selective media Chromocult® Tryptone Bile X-glucuronide agar (TBX) and Mannitol Salt Agar were used. Brilliance TM Salmonella Agar Base was used for *Salmonella* sp. detection; and Palcam Agar Base for *Listeria* sp. detection (Commission Regulation (EC) No 2073/2005 [31]). The number of yeasts was determined on Czapek Dox agar.

Serial dilutions were prepared from samples, and 0.1 mL from the homogenized sample mixture was spread on the surface of the respective selective media. The evaluated colonies were counted after 24 h of incubation at 37 °C [32].

2.7. Sensory evaluation

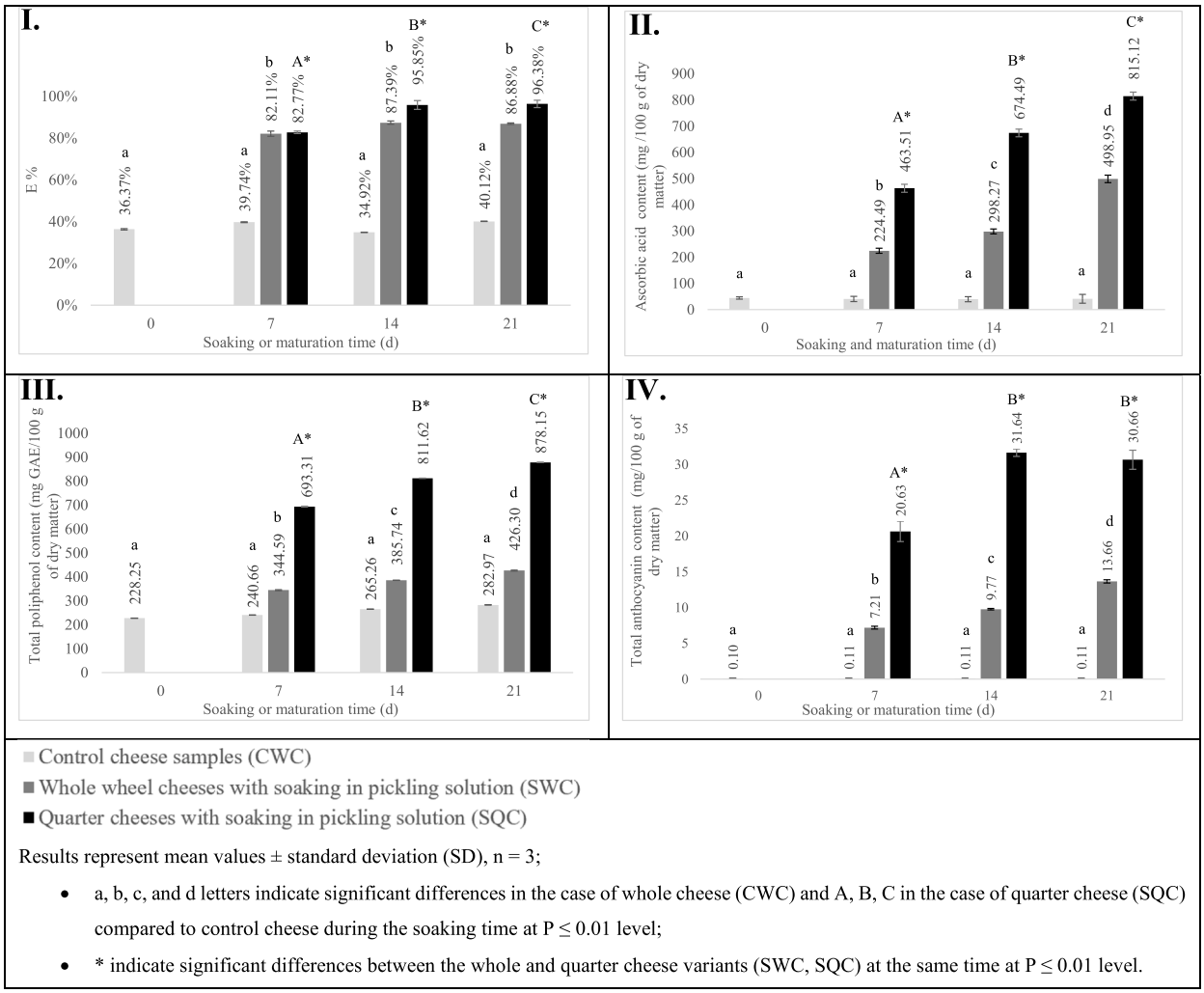
The assay was conducted based on the standard method SR 6345/1995 [33] guidelines published by the Romanian Standardization Association. In between sensorial analyses, apples were used for taste neutralization. The sensory analysis of cheese took place in the food laboratory at Sapientia Hungarian University of Transylvania, (Miercurea Ciuc, Romania) kitchen area, which we found to be compliant with SR 6345:1995 [33], with light-colored furniture, white walls, and a room free of foreign odors and noises. Throughout the tasting, there was natural light, without intense, strong sunlight, making it possible to evaluate the color of the cheese correctly [34].

For the sensory tests, a focus-group tasting method was used with 40 individuals (females 35 %, males 65 %) aged between 18 and 30. The samples of cheese were evaluated using a tasting questionnaire that was compiled to assess external appearance, color, appearance in the internal section, texture, aroma, and taste, in order to determine the characteristics of the cheeses in a uniform and transparent manner.

The samples were tasted at room temperature and cut into thin slices for proper consumption. The control sample was the whole cheese packed and stored at 4 °C in a vacuum bag (CWC).

The tasters evaluated the sensory characteristics based on a sensory evaluation questionnaire (supplementary file attached) with a 1–5 pointing scoring (1 - very bad, 5 - excellent) scale, which values were weighted by a constant: external appearance (0.4), color (0.4), appearance in its section (0.8), texture (0.4), smell (0.8), taste (1.2). According to the results of the sensory examination, the following categories were distinguished: excellent (20–18.1), good (18–15.1), sufficient (15–12.1), unsatisfactory (12–7.1), bad (7–4.1), very bad (4-0). In the case of dairy products, the minimum total score must be 12.1 (SR 6345:1995) [33].

Fig. 2. Evolution of dry matter content during the soaking process. Results represent mean values ± standard deviation (SD), n = 3; different letters (a, b, c) indicate significant differences from the control (CWC) at P ≤ 0.01 level; different letters (A, B) indicate significant differences between the variants (SWC, SQC) at P ≤ 0.01 level, * indicate significant differences between the whole and quarter cheese variants (SWC, SQC) at the same time at P ≤ 0.01 level.



Results represent mean values ± standard deviation (SD), n = 3;

- a, b, c, and d letters indicate significant differences in the case of whole cheese (CWC) and A, B, C in the case of quarter cheese (SQC) compared to control cheese during the soaking time at P ≤ 0.01 level;
- * indicate significant differences between the whole and quarter cheese variants (SWC, SQC) at the same time at P ≤ 0.01 level.

Fig. 3. Antioxidant activity properties of cheese: DPPH (I), FRAP (II), TPC (III) and TAC (IV).

2.8. Statistical analysis

For the statistical evaluation of chemical and microbiological data, one-way analysis of variance (ANOVA) was performed, for multiple comparisons the Tukey's HSD test was used, the differences between the means were considered statistically significant at P-values ≤ 0.01.

In the case of sensorial data statistical evaluation, Spearman's rank correlation analysis was used, performed in R software (version 4.2.1) P-values ≤ 0.05.

3. Results and discussion

3.1. Physicochemical characterization assays of cheese

3.1.1. Dry matter and fat content

The dry matter (DM) content of the cheeses was monitored from week to week to follow its changes over time and to measure to what extent the soaking and ripening time affect this parameter of the cheese. In the case of all examined samples of cheese, it can be seen (Fig. 2.) that with time, the DM content shows continuous increment, which was minor in the case of CWC (2.3 %) and higher in the case of SWC (12.7 %) and SQC (15.8 %) considering the three-week ripening and pickling period.

The biggest increment gap in DM content was observed after one week of ripening or soaking period: CWC: 1.3 %, SWC: 8.6 %, SQC: 13.6 %. The results show that by extending the ripening or soaking period, no significant change can be achieved regarding the DM content of the cheeses, the results are the same as those found in the literature [17].

The fat content of the ripened and soaked cheese was measured after 21 days. The results showed percentages of 29.89 % $\pm$ 0.2, 33.53 % $\pm$ 0.2, and 34.67 % $\pm$ 0.2 for CWC, SWC, and SQC, respectively. Comparing these values to the dry matter content of the cheese, it becomes apparent that soaking in pickling solution results in the release of a portion of the fat content due to the alcohol and salt content of the wine. The fat content for the control cheese is 46.63 % $\pm$ 0.2, whereas, for the soaked cheese, it measures 45.01 % $\pm$ 0.2 (SWC) and 44.68 % $\pm$ 0.2 (SQC).

3.1.2. Antioxidant activity results

The statistical evaluation of the data (Fig. 3.) clearly shows that while the soaking time in the pickling solution substantially affects the free radicals-scavenging ability of the SQC, in the case of the SWC, the maximum antioxidant activity is reached within two weeks and does not change significantly in the following week. It can be concluded that the DPPH assay of SQC increases significantly during the soaking time, so soaking for several weeks makes sense.

The difference between SWC and SQC in terms of DPPH, as seen in the case of TPC and FRAP is probably due to the higher specific surface area. So, if we want to produce a more valuable functional food, placing the cheese in a pickling solution in the form of slices would be the most efficient method.

During the ripening period, the free radicals-scavenging ability in the pickling solution does not vary significantly at $P \leq 0.01$ level (7th day 89.41 % $\pm$ 0.05, 14th day 88.98 % $\pm$ 0.15, 21st day 87.28 % $\pm$ 0.10).

Regarding ferric reducing antioxidant power (FRAP) analysis (Fig. 3.II.), the soaking in wine increased the antioxidant activity in both intact and sliced cheese samples. The weekly change increases similarly for both variants, the difference being that SQC shows almost twice as high FRAP value by the third week (815.12 mg AAS/100 g DM compared to the SWC value of 498.95 mg AAS/100 g DM).

Since polyphenols are primarily compounds that determine the coloring of fruits and vegetables, it is not surprising that their concentration in the case of the control cheese is very low in all periods of soaking (Fig. 3.III.). That small amount of TPC can also come from the cow's feed, partly excreted in the milk.

During the soaking experiment in the case of SWC, a constant linear growth can be experienced in the TPC, which increases by 40.86 ($\pm$0.3) mg GAE/100 g of DM from week to week. Accordingly, in the first week, the TPC of the cheese was 344.59 GAE/100 g DM; in the second week, this value increased to 385.74 GAE/100 g DM, and at the end of the soaking experiment, it reached a maximum value of 426.30 GAE/100 g DM. Furthermore, the rate of TPC increase during the experiment is even more pronounced in the case of SQC, which can be attributed to the higher specific surface area of SQC compared to SWC. In the first week, the TPC of SQC was more than 100 % higher, compared to SWC, namely 693.31 GAE/100 g DM, which increased to 811.62 GAE/100 g DM by the end of the second week, and finally, the TPC reached a value of 878.15 GAE/100 g DM at the end of the third week. It is evident from the obtained values that the growth rate in TPC is even higher than expected from the increased specific surface. The specific area of SQC is 177.3 % (height: 5 cm, diameter: 8 cm) higher compared to that of the SWC, but the TPC is more than twice as high at each of the three measurement times. At the same time, it should also be noted that further shredding the cheese during soaking might have a positive effect on the phenol content (as a functional food component). However, due to its ethanol content the marinade could extract more fat and other non-polar compounds from the cheese, which would confer upon the cheese a dry effect which, on its turn, would lower the consumer's satisfaction. Cavalcanti et al. [35] studied changes in total polyphenol content in goat cheese soaked in red wine. They soaked the cheese for seven days and measured a similar proportion of difference to the control cheese as we did.

Looking at the total anthocyanin content, while the anthocyanin content of SWC increases steadily over the three weeks, SQC reaches a maximum in the second week, with little or no change by the third week (Fig. 3.IV.).

Table 1
Microbiological load of cheese samples over time.

Cheese name, sampling date	*E. coli*	*S. aureus*	Yeasts	Lactic acid bacteria
	CFU/g [mean ± (SD)]			
CWC, day 0	$1.90{\cdot}10^{3a}$ ($\pm 4.91{\cdot}10^{1}$)	$1.20{\cdot}10^{3a}$ ($\pm 6.22{\cdot}10^{1}$)	$4.20{\cdot}10^{2a}$ ($\pm 1.11{\cdot}10^{1}$)	$2.14{\cdot}10^{6a}$ ($\pm 2.15{\cdot}10^{5}$)
CWC, day 7	ND * ^{b}	$7.00{\cdot}10^{1b}$ ($\pm 3.51{\cdot}10^{0}$)	ND^{b}	$5.75{\cdot}10^{7b}$ ($\pm 2.18{\cdot}10^{6}$)
SWC, day 7	ND ^{bA}	$1.00{\cdot}10^{2}$ ^{cA} ($\pm 5.13{\cdot}10^{0}$)	$8.50{\cdot}10^{2}$ ^{cA} ($\pm 1.50{\cdot}10^{1}$)	$6.59{\cdot}10^{7}$ ^{cA} ($\pm 4.69{\cdot}10^{6}$)
SQC, day 7	ND ^{bA}	$7.00{\cdot}10^{1}$ ^{dA} ($\pm 6.11{\cdot}10^{0}$)	$9.00{\cdot}10^{1}$ ^{dB} ($\pm 5.13{\cdot}10^{0}$)	$4.27{\cdot}10^{6}$ ^{aB} ($\pm 1.54{\cdot}10^{5}$)
CWC, day 21	ND^{b}	ND^{e}	$4.00{\cdot}10^{2e}$ ($\pm 1.39{\cdot}10^{1}$)	$4.81{\cdot}10^{7d}$ ($\pm 4.22{\cdot}10^{6}$)
SWC, day 21	ND ^{bA}	ND ^{eB}	$2.00{\cdot}10^{1}$ ^{fC} ($\pm 3.51{\cdot}10^{0}$)	$1.65{\cdot}10^{5}$ ^{eC} ($\pm 8.84{\cdot}10^{3}$)
SQC, day 21	ND ^{bA}	ND ^{eB}	ND ^{bD}	$1.75{\cdot}10^{4}$ ^{fD} ($\pm 9.13{\cdot}10^{2}$)

Results represent mean values ± standard deviation (SD), n = 3; different letters (a, b, c, d, e, f) indicate significant differences within a microorganism or group of microorganisms from the control on day 0 (CWC, day 0) at $P \leq 0.01$ level; different capital letters (A, B, C, D) indicate significant differences between the variants (SWC, SQC) at the same sampling day within a microorganism or group of microorganisms at $P \leq 0.01$ level; * - not detected.
E. coli and *S. aureus* were present in the initial samples at $1.90{\cdot}10^{3}$ CFU/g and $1.20{\cdot}10^{3}$ CFU/g, but they were no longer detectable at the last sampling time (day 21).

3.1.3. Ethanol content of soaked cheese

The ethanol content of the cheese was measured once at the end of the experiment (after 21 days of soaking) by HPLC, and the values were expressed as g ethanol/100 g cheese. According to this, in the case of SWC, the ethanol content was 0.43 ± 0.02 g/100 g cheese, while in the case of SQC, this value was 1.29 ± 0.08 g/100 g. Due to the low alcohol content of the cheese, it is safe for consumption by healthy adults.

3.2. Microbiological assay

The microbiological assay of the salted blackcurrant wine (5 % w/V) during soaking (at day 0, at the end of 1st week, and at the end of 3rd week) resulted that no yeasts were detected, indicating that the salted (Commission Regulation (EC) No 2073/2005 [31]) wine's yeast contamination was below the detection range. In the case of the lactic acid bacteria group, only 1 CFU/mL was detected in the sample taken from the wine at day 0. In red wines, lactic acid bacteria contribute to maleic acid metabolism.

After the cheese samples were obtained and soaked in salted blackcurrant wine, a control cheese was placed in vacuum foil for ripening at the same temperature. The microbiological quality of the samples of cheese is summarized in Table 1.

In the soaked cheese samples, *E. coli* was absent both in the whole and quarter cheese samples, while in the case of *S. aureus*, on day 7, there were detected a quantity of 100 CFU/g SWC and 70 CFU/g SQC samples, which is not considered a significant difference between the variants. After three weeks, none of these foodborne pathogens were detectable in the cheese samples. This results from the fact that salted blackcurrant wine has an antimicrobial effect due to its alcohol and salt content.

One of the hygienic indicator microorganisms during cheese production is *E. coli*. The presence of this bacteria reflects fecal contamination. These bacteria exhibit diverse groups based on phenotypic characteristics and specific virulence factors, potentially leading to health issues [32,36].

S. aureus was detected in some dairy products as a predominant zoonotic pathogen. Also, it is associated with the most common cause of foodborne outbreaks. Good hygiene and suitable acidification conditions are recommended to prevent the multiplication of *S. aureus* during cheese production [32,37].

The presence of yeast was detectable even after 21 days. In cheesemaking, yeasts are considered part of secondary microbiota and depend on physicochemical conditions. Some species are considered contaminants responsible for different cheese defects [38,39]. The spoilage yeasts also could be present in wine, surviving the wine environment conditions such as low pH, high ethanol concentration, the concentration of sulfites and dimethyl dicarbonate [40].

The lactic acid bacteria cell count was continuously present over time, ranging from $6.59{\cdot}10^{7}$ CFU/g to $1.75{\cdot}10^{4}$ CFU/g; their number showed a significant decrease in the case of SWC. The results showed that the number of lactic acid bacteria in the cheese samples decreased depending on soaking time, i.e., blackcurrant wine has an inhibitory effect, but further studies are needed to understand the mechanism involved. During the 7 days of soaking, the smallest change in the number of lactic acid bacteria was observed in the SWC. To prevent the inhibition of the activity of lactic acid bacteria, a maximum soaking time of 7 days is recommended for both SWC and SQC cheeses. After 7 days of soaking, we observed a significant increase in the number of lactic acid bacteria in both the control (CWC) and SWC cases. Within the same time frame, a slight increase in cell count was observed in the case of SQC; however, this could not be deemed a significant elevation. However, following a 21-day soaking period, both SWC and SQC exhibited a significant decrease, ranging from one to two orders of magnitude, in the count of lactic acid bacteria.

The number and the diversity of lactic acid bacteria represent an impact on cheese quality and type. Cheese is produced using lactic acid bacteria as added starter cultures or non-starter lactic acid bacteria. Their major role is the lactose transformation and, with other biochemical changes during ripening, they contribute to the characteristic organoleptic properties of the cheeses or the safety of the product [41,42]. It was assumed that lactic acid bacteria in fermented products would contribute to the dynamics of the formation of volatile compounds [34]. *Salmonella* sp. and *Listeria* sp. were not detected in any of the samples. The antibacterial activity of alcohol-containing soaking solution against *Salmonella* sp. was previously described [35].

Table 2
The acceptance test results.

Properties	Weighted average score values of		
	CWC	SWC	SQC
External appearance	2.00 (±0.12)	1.98 (±0.09)	1.92 (±0.16)
Color	1.98 (±0.14)	1.92 (±0.16)	1.92 (±0.16)
Appearance in its section	3.88 (±0.58)	3.48 (±0.70)	3.56 (±0.61)
Texture	1.98 (±0.34)	1.95 (±0.36)	1.77 (±0.38)
Smell	3.88 (±0.51)	3.8 (±0.44)	3.76 (±0.61)
Taste	5.88 (±0.68)	5.82 (±0.89)	5.46 (±0.78)
Sum	***19.6***	***18.95***	***18.39***
Rating category	Good	Good	Good

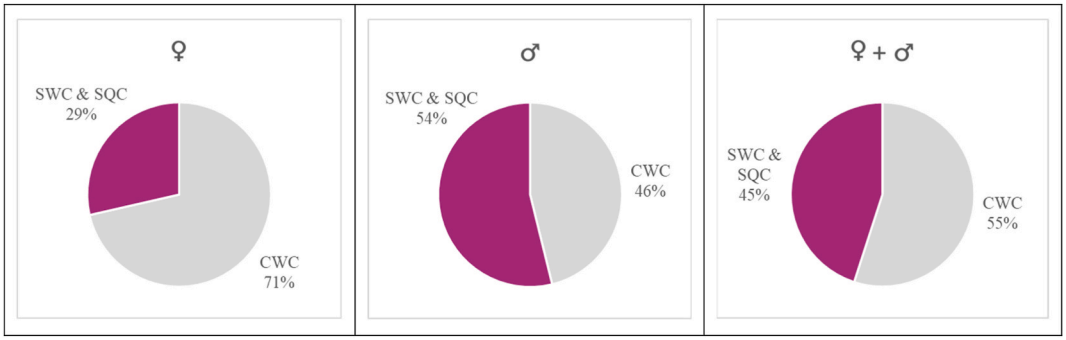


Fig. 4. Gender distribution of sensory test results.

Based on the microbiological results, all samples indicate that they meet the criteria for consumption and market placement without any restrictions, in accordance with the European Union's regulations on effective microbiological limit values (Commission Regulation (EC) No 1441/2007 [43]).

3.3. Sensory evaluation

The whole and quarter samples of cheese soaked in the pickling solution (SWC, SQC) were monitored week by week, both their appearance and their internal texture, to see how well the wine was able to diffuse into the surface of the cheeses and how the color of the cheese changed during the soaking period. The evolution of the soaking of SWC and SQC samples after one week, two weeks, and three weeks are shown in appendix 1. After one week of soaking in the pickling solution, the outer surface of the cheese was dark red color of the blackcurrant wine, which gave the cheese a beautiful appearance, making it even more special. During this short period, the wine penetrated the cheese in a very thin layer, so the interior of the cheese retained its characteristic slightly yellowish color. We removed a cheese set from the pickling solution at the end of the second week. The color of the cheese changed from dark reddish color to lighter purple. Regarding the internal stock of cheese, albeit minimally, the thickness of the crust increased, and the inside of the cheese became slightly yellower due to the ripening process. After three weeks of ripening in the pickling solution, the color of the cheese was rather dark purple, and the inside color of the cheese changed to the characteristic yellow color of ripened cheese.

Forty participants evaluated the cheese in the sensory analysis and completed the tasting questionnaire. The participants were aged 23–35 years. In terms of the gender distribution of the participants, 35 % were female, and 65 % were male. 65 % of participants had not previously participated in similar sensory assessments. All tasters consume cheese weekly; for 35 % cheese is part of their daily diet.

Based on the scores gained after 3 weeks of soaking (Table 2), both categories of cheese, namely the whole and quarter cheese, ripened in a pickling solution, and the control cheese were classified as good, so they fit it in the *good category*.

The SWC and SQC scored a total of 18.95 and 18.39 points, respectively, compared to the CWC's 19.6 points. It can be seen that the difference between the scores was mainly determined by appearance, texture, and taste. This difference is likely attributed to the fact that the SWC and SQC samples' appearance and taste are different from the usual ones, which, for some people, was exciting and special. In contrast, for others, it was not really confidence-inspiring. Regarding the texture of the cheese, the texture of CWS was creamier, softer, and silkier, while the texture of SWC and SQC had a harder and drier effect, which was also remarked by the tasters when scoring. The color of the SWC and SQC, as well as the taste, were also quite different from the control cheese: some evaluators liked the characteristic reddish-purple color, but some did not find it appetizing and preferred the regular, slightly yellow cheese. The tasters were asked which type of cheese they would prefer to eat out of the examined samples of cheese. Taking gender into account, the results are presented in Fig. 4.

Overall, 55 % of the respondents preferred the traditional ripened cheese, while 45 % preferred the soaked cheese. Among the tasters, men said they would regularly consume cheese ripened in the pickling solution, whereas women choose regular, traditionally

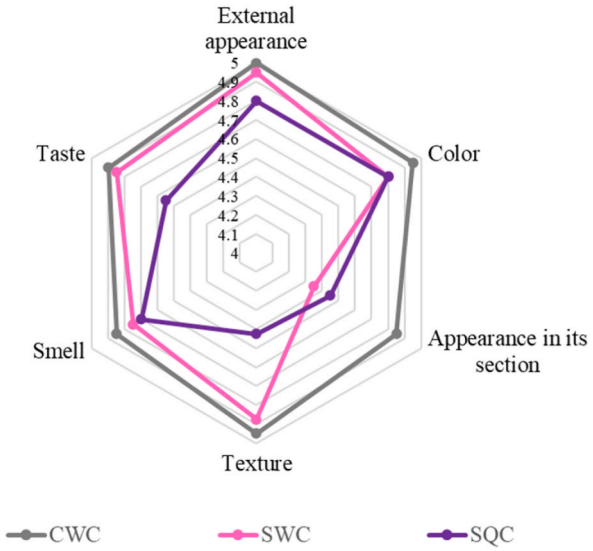


Fig. 5. Results (expressed as the median) of the sensory analysis for cheese samples.

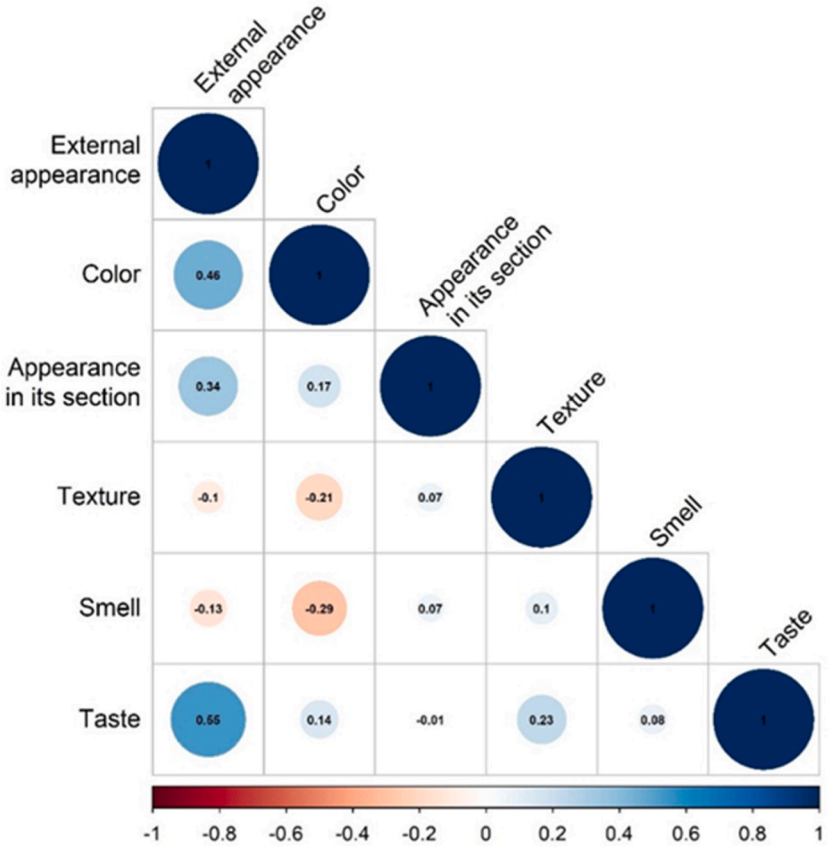


Fig. 6. Spearman's rank correlation of the sensory parameters for SWC cheese samples.

ripened cheese. It can be assumed that the difference in opinion regarding the consumption of cheese between the sexes can be attributed to the fact that the cheese ripened in the pickling solution may have not only the pleasant fruity taste of blackcurrant but also the typical wine taste, which can induce a more favorable and pleasant feeling in men.

The average scores the reviewers gave for analyzed sensorial properties of cheese are summarized in Fig. 5.

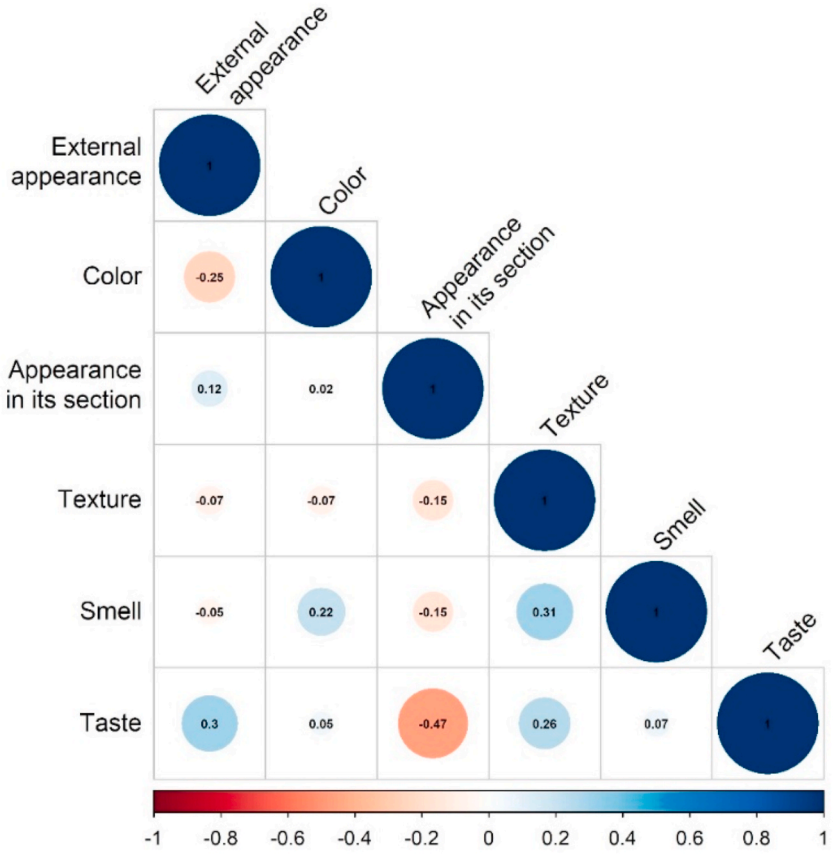


Fig. 7. Spearman's rank correlation of the sensory parameters for SQC samples. A negative correlation in the case of SWC (Fig. 7.) was found between the color (r = −0.25), smell (r = −0.15), texture (r = −0.15).

Analyzing the sensory properties of the samples (Fig. 5.) showed the highest score for the control cheese samples, followed by the scores of the whole soaked cheese. SWC samples were valued with high scores for external appearance, taste, smell and texture. The significant difference between the two soaked cheese samples was the texture.

Based on the number of evaluators (N = 40, P < 0.05), the significance level was set at r = 0.38. Given this significance level, in the case of SWC a positive correlation was found between taste (r = 0.56) and color (r = 0.46) (Fig. 6).

4. Conclusions

The soaking of cheese has a substantial impact only in the first week, which manifests in the growth of the dry matter of the cheese. From the point of view of increasing the antioxidant activity, it is advisable to soak quartered cheeses and preferably soaked for a minimum of seven days. After three weeks of cheese soaking, the antioxidant content of the pickling solution decreased only very slightly, so the cheese-to-pickling solution ratio could be reduced to maintain the original ratio; however, further experiments are needed in this respect. Soaking in the pickling solution benefits cheeses, as it stops the growth of bacteria that cause foodborne illnesses. Cheese ripened in salted blackcurrant wine is suitable for consumption owing to its sensory and physicochemical properties. Due to the lactic acid fermentation, it possesses enhanced nutritional value. Although the soaked cheese has a low alcohol content, it is recommended for consumption, suggested primarily for adults. Blackcurrant wine-soaked cheese could be a valuable and feasible way to improve sensorial characteristics, functionality, and food safety.

Ethical statements

Ethical approval and consent to participate

The authors declare that they have the consent of the Bioethics Committee of Sapientia Hungarian University of Transylvania (approval number 543/October 30, 2023), and all participants consented to take part in the sensory evaluation of the product and use their information. All procedures were performed in compliance with relevant laws and institutional guidelines. The appropriate protocols for protecting the rights and privacy of all participants were utilized during the execution of the research.

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Data availability statement

The data that support this study are available from the corresponding author upon reasonable request.

Additional information

No additional information is available for this paper.

CRediT authorship contribution statement

László Gyenge: Writing – review & editing, Writing – original draft, Software, Formal analysis, Data curation. **Kinga Erdő:** Writing – original draft, Investigation, Data curation. **Csilla Albert:** Supervision, Methodology, Investigation. **Éva Laslo:** Writing – original draft, Methodology, Data curation. **Rozália-Veronika Salamon:** Writing – review & editing, Validation, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Conceptualization.

Declaration of competing interest

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

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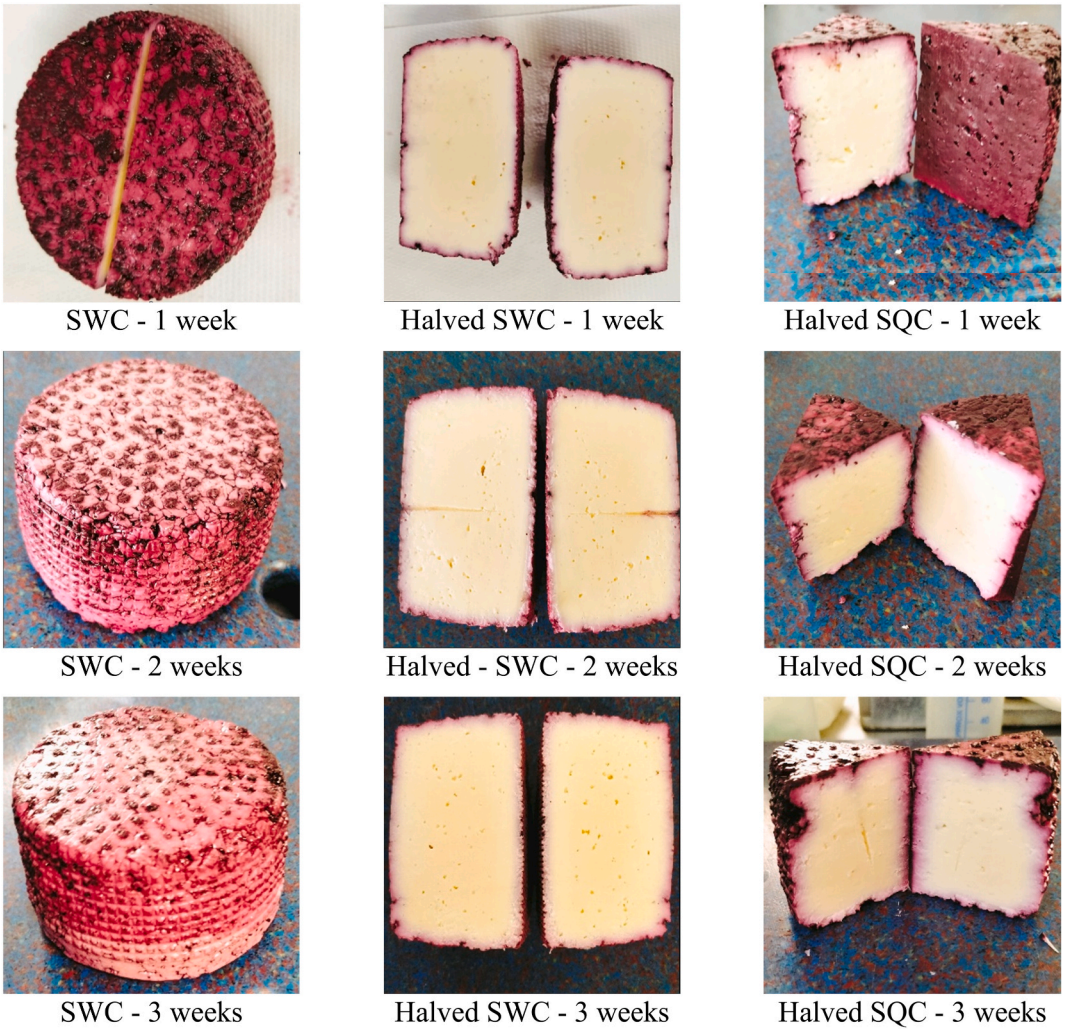
Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e34060.

Appendix



Appendix 1. The color of whole and quarter-soaked cheese over the soaking and ripening process.

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