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The effect of nisin and storage temperature on the quality parameters of processed cheese

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Abstract

The aim of this study was to evaluate the effect of nisin and storage temperature on the microbiological, physico-chemical and sensory properties of processed cheese. Processed cheese samples were prepared without and with 12.5 ppm nisin, stored at different temperatures (4, 8 and 20 °C). Research covered microbiological, physico-chemical and sensorial analysis of the samples. In processed cheese samples prepared without nisin, aerobic colony count (ACC) significantly increased over the experimental period, while in nisin supplemented samples no significant increase was determined. At lower storage temperature (4 °C), the effectiveness of nisin was more pronounced. Yeast and moulds (YM) and psychrotrophic colony count (PCC) were below the limit of quantification over the entire experimental period. Also, no significant changes in pH values and textural characteristics (firmness and stickiness) were determined. Sensory analyses confirmed that processed cheese samples became whiter and less yellow, compared to fresh samples. The viscosity of all samples decreased over the experimental period, which was in line with the results of baking stability. The obtained results confirmed the potential of using this type of processed cheese in baking industry.

Key words: processed cheese, quality parameters, nisin, storage temperature

Introduction

Processed cheeses are cheese-based products produced by blending natural cheeses of different types and degrees of maturity and emulsifying agents. The mixture is heated until a homogenous blend is obtained (Kapoor and Metzger, 2008). In addition to natural cheeses, other dairy and non-dairy ingredients may be included in the blend, such as fat, milk powder, whey proteins, caseins, water, vegetable fat, colorants, flavourings, salts, hydrocolloids, etc. These products usually have 40-60 % water content. The pH values are in the range from 4.1 to 6.0, while water activity (a_w) values fall between 0.93 and 0.95. Usually, the processing of this type of cheese involves heat treatment at a temperature of 80-100 °C, for 5-15 min. Although this thermal treatment is appropriate for destroying vegetative forms of present microflora, it will not be sufficient to kill bacterial spores. Therefore, processed cheese

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may contain viable spores, which originate from natural cheese or other added ingredients. This type of contamination in processed cheese originates from gram-positive spore-forming bacteria of the genus *Bacillus* and *Clostridium* (Glass and Doyle, 2013; Glass and Johnson, 2004; Oliveira et al., 2016). Therefore, the microbiological quality of processed cheeses is dependent on the quality of ingredients, as well as on other factors such as pH, moisture, fat, salts and/or the presence of food additives (Glass and Johnson, 2004). Literature data indicated that these products are considered as safe. During the long production history of almost 40 years, only few outbreaks occurred, involving products with high pH (>5.7) and a_w (>0.96) values (Anderson et al., 2011).

One of the most extensively studied bacteriocin is nisin, which has gained widespread application in dairy industry (de Arauz et al., 2009; Ávila et al., 2014). It is produced by certain strains of *Lactococcus lactis* and has an inhibitory effect on the growth of a number of gram-positive organisms, including species in the genera *Clostridia*, *Bacillus*, *Lactobacillus*, *Lactococcus*, *Micrococcus*, *Listeria*, *Staphylococcus*. When applied in combination with thermal processing, other bacteriocins, high pressure, low-temperature storage, reduced pH and/or a_w, nisin can prolong shelf-life and enhance food product safety and quality (Oliveira et al., 2016). This type of application is considered as the hurdle concept (Rajkovic et al., 2010; Smigic and Rajkovic, 2014). Finally, the usage of 12.5 ppm of nisin in processed cheese is legally approved (EU, 2008; Serbia, 2013).

Spoilage of processed cheese might occur due to the microbial growth, and is usually detected by the gas production, abnormal odours/colours or pH variations. As a consequence of these changes, various sensory defects could occur, such as off-odour, discoloration, unusual texture properties, etc. (Ledenbach and Marshall, 2009). In addition, various chemical and physical changes may impair sensorial characteristics of processed cheeses (Schär and Bosset, 2002). The textural properties, including firmness, stickiness and adhesiveness, but also the other quality descriptors of processed cheeses are important depending on their purpose. Viscosity and baking stability are affected by various factors, such as the shelf-life and composition of natural cheeses, the pH of the blend, the processing time and temperature, the storage conditions, presence and concentration of calcium, sodium and potassium ions, type and concentration of sugars and emulsifiers used (Silva et al., 2012). Processed cheeses are suitable for the food industry, as they can be tailored for specific food sectors, such as the baking industry.

The aim of this study was to evaluate the effect of nisin (12.5 ppm) and storage temperature (4, 8 and 20 °C) on the microbiological, physico-chemical and sensorial quality of processed cheese.

Materials and methods

Processed cheese production

For the purpose of this experiment, processed cheese samples were prepared, using the following ingredients: whey cheese, water, modified corn starch, dextrose, salt, sodium citrate, lactic acid, yeast extract and potassium sorbate. Initially, all ingredients were blended with water, homogenized and subsequently processed by heating and shearing. Processing was carried at the temperature 90 °C for 15 min in Vorwerk Thermomix (Wuppertal, Germany). Two batches of samples were prepared, with nisin at the concentration of 12.5 ppm (Danisco, Denmark), and without nisin. Samples were packaged in 1 kg packages, closed with foil and tempered at room temperature for 24 h. Afterwards samples were stored at different temperatures, namely 4, 8 and 20 °C. The complete experiment was performed in three repetitions.

Microbiological analysis

The aerobic colony count (ACC), psychrotrophic colony count (PCC), yeasts and moulds (YM) and *Enterobactericeae* in processed cheese samples were enumerated. The 10 g sample was mixed into 90 mL of sterile Physiological Peptone Solution (PPS, Oxoid) in sterile stomacher bag and homogenized for 2 min. Decimals dilutions were prepared and plated on specific media.

ACC were enumerated by the pour plate method using Plate Count Agar (PCA) (Torlak, Serbia), and incubated at 30 °C for 48 h. PCC were determined on PCA (Torlak, Serbia) by the pour plate method and subsequent incubation at 8 °C for 10 days. Yeast and moulds were determined using DG18 agar supplemented with glycine (Centrohem, Serbia) after the incubation at 25 °C for 3-5 days. *Enterobactericeae* were determined using Violet Red Bile Glucose Agar (VRBG) (Oxoid, UK), and subsequent incubation at 37 °C for 24 h. Obtained results were expressed as log CFU/g. Samples that were stored at 4 °C and 8 °C were analysed every second week during the period of 12 weeks, while samples stored at 20 °C were analysed every second week during 8 weeks.

Compositional analysis

Physico-chemical analysis of processed cheese samples were performed in the first week after the production, according to the following methods: dry matter by standard drying method at 102±2 °C (FIL-IDF, 1982); total protein content by analyzing nitrogen content with Kjeldahl method (AOAC, 1990) and multiplying it with 6.38. Water activity was determined using A_w-meter (Testo, model 650, Germany). The pH of all processed cheese was measured every second week during the storage time of 8 weeks. For that purpose, 10 g of sample was homogenized with 10 mL of distilled water and subsequently pH was determined using a digital pH-meter (Consort, Turnhout, Belgium).

Physico-chemical analysis

Baking stability test

Baking stability test was used to determine the heat-stability properties of the processed cheese samples, as these samples might be used for the pastry fillings in the baking industry (Cropotova et al., 2013; Young et al., 2003). For that purposes a certain amount of prepared sample was placed on the filter paper within a metal ring (50 mm diameter and 10 mm height). Samples were heated at 190 °C for 10 min. Pictures of the samples were taken before and after the baking, using Sony Alpha 100 camera (Sony, Tokyo, Japan). Obtained images were analysed using program ImageJ software 1.38v to determine the surface changes of the samples. Results were presented as the % baking stability (BS) using formula (1):

$$BS\ (\%) = \frac{D_2 - D_1}{D_1} * 100 \qquad (1)$$

where:

BS (%) - baking stability,

D_1 - surface area of the sample before baking, cm^2,

D_2 - surface area of the sample after baking, cm^2.

Samples were analysed every second week during storage time of 8 weeks.

Viscosity

To determine viscosity, samples were placed in a programmable Fungilab S.A. V 1.2 Alpha Series viscometer at 25 °C (Fungilab, Spain). The viscosity of samples was measured every second week during storage time of 8 weeks.

Texture profile analysis

Textural parameters, firmness and stickiness, were measured by the TA.XT Plus Texture Analyzer (Stable Micro System, Godalming, Surrey, UK) equipped with spread ability ring (HDP/SR), heavy duty platform (HDP/90), male and female cones. After calibration, the following parameters were adjusted for the measurement test speed - 3 mm/s; penetration depth - 15 mm and trigger force - 2 g. For data analysis, Texture Exponent program was used. Samples were analysed every second week during storage time of 8 weeks and each measurement was performed in five repetitions.

Colour measurement

Visual colour of processed cheese samples was measured based on CIELab D_{50} colour parameters using computer vision system (CVS) identical to the one used in the work of Smigic et al. (2017).

The total colour change ΔE was calculated using the formula (2):

$$\Delta E = \sqrt{(\Delta L^2 + \Delta a^2 + \Delta b^2} = \\ = \sqrt{(L - L_{initial})^2 + (a - a_{inital})^2 + (b - b_{initial})^2} \qquad (2)$$

where:

ΔL- difference at a certain point compared to the day of sample preparation ($L_{initial}$) in the dark-bright scale,

Δa- difference at a certain point compared to the day of sample preparation ($a_{initial}$) in the red-green scale,

Δb- difference at a certain point compared to the day of sample preparation ($b_{initial}$) in the yellow-blue axis.

Colour changes were expressed as whiteness index (WI) and yellowness index (YI) using the following formulas (Ramirez-Navas and Rodriguez de Stouvenel, 2012; Oms-Oliu et al., 2008):

$$WI=100-\sqrt{(100-L^*)^2+(a^*)^2+(b^*)^2} \quad (3)$$

$$YI=142.86*\frac{b^*}{L^*} \quad (4)$$

Samples were analysed every second week during storage time. Initial values were obtained for the fresh sample analysed after preparation (first week).

Sensory analysis

The changes of sensorial parameters over time, can be estimated by measuring the degree of difference between stored samples and a "control" sample, using intensity scales (Giménez et al., 2012). For each sensorial analysis, a freshly prepared and 1 week stored processed cheese sample was appointed as a "control" sample. Samples were evaluated by a trained seven-member panel, consisting of researchers from the Department of Animal origin food and Department of Food Safety and Quality Management. Analyses were carried in the Laboratory for Sensory analysis, under day light. Samples stored at 4 °C and 8 °C were left at room temperature two hours before sensory testing. All samples were presented in plastic trays. The sensory analysis was performed in two replications. Samples together with a "control" sample were presented to the panellists in a random order. Each sample was coded with a randomly selected 3-digit number. Panellists were instructed to cleanse their palates between samples using water. Samples stored at 4 °C and 8 °C were analysed every second week during 12 weeks, while samples stored at 20 °C were analysed every second week during 8 weeks.

Samples were compared with the "control" sample for the following attributes: colour, taste and odour. An intensity scale test of 9-points was used for each attribute: colour (1=too light, 5=control, 9=too dark), taste (1=not enough taste, 5=control, 9=too much taste); odour (1=not enough odour, 5=control, 9=too much odour). In addition, overall appearance defects, if any, were evaluated using a 5-point intensity scale (5=control; 9=very intense defects). Schematic differential chart was used to visualize the sensory profile of the samples.

Data analysis

Obtained data were analysed using analysis of variance (ANOVA) with post hoc Duncan test to distinguish statistical differences between different processed cheese samples and same samples over time. Values with $p<0.05$ were considered statistically significant. All statistical processing was performed using SPSS Statistics 17.0.

Results and discussion

Microbiological analysis

The results of aerobic colony count (ACC) are presented in Table 1. ACC significantly increased in processed cheese samples prepared without nisin ($p<0.05$), while in nisin supplemented samples no significant increase was determined ($p>0.05$). This might be explained by the known antimicrobial activity of nisin that inhibited the growth of present microbial flora. Zottola et al. (1994) also reported that cheese spreads containing 300-390 IU nisin/g had a significantly longer shelf life, compared to control samples without nisin. They have followed the cheese spread samples inoculated with *Clostridium sporogenes* at 22 °C, and obtained results indicated that nisin delayed spoilage to 87 days, compared to 14 days for samples without nisin. In addition, results of their study suggested that the control of food-borne pathogens *L. monocytogenes* and *S. aureus* could be obtained by using a nisin containing cheese in cheese spread formulation. The study of Somers and Taylor (1987) also showed that addition of nisin in the preparation of the pasteurised cheese spread prevented the botulinum growth and toxin production. Their results indicated that the cheese spreads formulated with 52 % moisture, pH 5.9 and 2.5 % disodium phosphate require only 12.5 ppm nisin to inhibit toxin production. Our results also indicated that ACC in all analysed samples was in the range of 2.14 - 2.68 log CFU/g at the end of the experimental period, being far below an unacceptable level (>6 log CFU/g) (Nicoli, 2012; Dermiki et al., 2008; Bolton et al., 2009).

Along with the ACC, psychrotrophic colony count (PCC) was also determined. Except for three single measurements, namely, samples stored at 4 °C, with nisin (2.23 and 3.50 log CFU/g) and without nisin (2.31 log CFU/g), in all other samples, PCC was below the limit of quantification (being 1.7 log CFU/g, data not shown).

Similarly, in only three samples, number of yeast and moulds (YM) was above the limit of quantification (being 2.7 log CFU/g, data not shown). These single measurements (not mean values) were determined for samples stored at 20 °C with either added nisin (3.62 and 4.22 log CFU/g) or without nisin (5.53 log CFU/g). The possible explanation for these "outliers" could be the contamination from the air during manual weighting and packaging of the processed cheese samples. As this is not seen in other samples and in all replicates, it might indicate only occasional contamination and outgrowth.

YM or PCC may cause cheese products spoilage when present in great number (>6-7 log CFU/g) (Pereira-Dias et al., 2000; Fleet, 1990). Our results confirmed no spoilage within the observed period, which was also confirmed through sensory analysis.

Compositional analysis

The obtained results for compositional analysis indicated that the content of dry matter was 30.74±0.23 %, the total protein content was 6.56±0.04 %, while water activity was a_w=0.95±0.004. In line with these results in freshly prepared samples (week 1), pH was analysed during the entire experimental period to determine, if significant changes will occur. The obtained results are presented in Table 2. As it can be seen, all values were in the range between pH=4.99-5.06, which indicated relative stability of the product. These results were in line with microbiological results, as no important increase of either ACC or PCC was seen, whose growth might result in pH changes (Lycken and Borch, 2006). In addition, the solubility of nisin is pH dependent, and therefore the production of low pH stable product would emphasise the bacteriocin activity (Zottola et al., 1994).

Table 1. Aerobic colony count (ACC) for processed cheese samples (log CFU/g) stored at different storage temperature (4 °C, 8 °C and 20 °C) in samples with nisin (12.5 ppm) and without nisin during 12 weeks of storage. Values represent mean ± standard deviation (n=3)

	Aerobic colony count (ACC) (log CFU/g)					
	Storage temperature, 4 °C		Storage temperature, 8 °C		Storage temperature, 20 °C	
Time (weeks)	With nisin	Without nisin	With nisin	Without nisin	With nisin	Without nisin
1	2.16±0.05^{AB}	2.13±0.05^{aAB}	2.08±0.17^{A}	2.22±0.15^{aAB}	2.09±0.23^{A}	2.36±0.02^{aB}
3	2.16±0.23	2.28±0.17^{ab}	2.16±0.70	2.24±0.14^{a}	1.94±0.25	2.13±0.16^{a}
5	1.80±0.43^{A}	2.40±0.10^{bB}	1.94±0.32^{AB}	2.35±0.12^{abAB}	2.07±0.33^{AB}	2.33±0.25^{aAB}
7	2.08±0.15^{AB}	2.33±0.14^{bB}	2.12±0.09^{AB}	2.38±0.04^{abB}	1.86±0.36^{A}	2.81±0.23^{bC}
9	1.80±0.17^{A}	2.34±0.07^{bB}	2.09±0.43^{AB}	2.49±0.11^{bcB}	ND*	ND*
11	1.86±0.13^{A}	2.43±0.06^{bB}	2.04±0.16^{A}	2.36±0.04^{abB}	ND*	ND*
12	2.14±0.03^{A}	2.44±0.04^{bB}	2.17±0.01^{A}	2.68±0.09^{cC}	ND*	ND*

Means in the same column with different small letters and means in the same row with different capital letters are significantly different ($p<0.05$)
*ND - not determined

Table 2. pH values determined in different processed cheese samples stored at different temperature (4 °C, 8 °C and 20 °C) and prepared without and with nisin (12.5 ppm). Values represent mean ± standard deviation (n=3)

	pH values, mean±standard deviation (n=3)					
	Storage temperature 4 °C		Storage temperature 8 °C		Storage temperature 20 °C	
Time (weeks)	With nisin	Without nisin	With nisin	Without nisin	With nisin	Without nisin
1	5.06±0.01^{aC}	5.05±0.01^{BC}	5.05±0.01^{BC}	5.02±0.00^{abAB}	5.00±0.04^{A}	5.01±0.03^{AB}
3	5.00±0.01^{bA}	5.03±0.04^{AB}	5.03±0.01^{AB}	5.05±0.01^{abB}	5.02±0.01^{AB}	5.02±0.01^{AB}
5	4.99±0.01^{bA}	5.00±0.01^{AB}	5.06±0.02^{C}	4.98±0.04^{bA}	5.04±0.02^{BC}	5.05±0.04^{BC}
7	5.02±0.04^{b}	5.01±0.05	5.04±0.02	4.99±0.05^{b}	5.01±0.04	5.01±0.03

Means in the same column with different small letters and means in the same row with different capital letters are significantly different ($p<0.05$)
*ND - not determined

Physico chemical analysis

Baking stability test

Processed cheese samples can be used for the fillings in the savoury baked products, and therefore it was important to determine if the baking process itself would affect their stability. For that purposes, baking stability test was performed (Cropotova et al., 2013; Young et al., 2003) and obtained results are presented in Figure 1. It is obvious that during the storage time this parameter changed. At the beginning, baking stability index was negative in all samples, indicating that the surface of the samples was smaller after baking than before baking. Nevertheless, during storage time this parameter changed in different direction and values for baking index were positive, which means that the surface of the samples was greater after baking than before baking process. Possible explanation is that samples stored for a longer period of time at a given temperature, have less strength and weaker structure obtained by modified corn starch (Saha and Bhattacharya, 2010).

Viscosity

Different chemical and compositional factors may affect functional properties of processed cheese, such as viscosity. Nevertheless, the goal of this study was not to identify influence of different parameters on the values of viscosity, but rather to determine the changes during storage time in samples with and without added nisin. The obtained results are presented in Figure 2. It is obvious that there is no significant difference between different samples ($p>0.05$). Therefore no influence of nisin or storage temperature could be seen. It is however important to note that relatively low viscosity values have been determined in this study. This might be explained by the lower protein content in the samples compared to processed cheese samples reported elsewhere (Sołowiej et al., 2014), as low protein content may result in poorer viscosity (Sołowiej et al., 2014). The results presented in Figure 2 showed that the viscosity of all samples decreased during the storage time. At the first week of storage, values were in the range 130-144 Pa·s, compared to the range 109-122 Pa·s at the end of observation period. Modified starches are usually used to thicken the complete blend for processed cheese preparations, but they also increase the viscosity of the blend (Wüstenberg, 2015). Therefore, at the beginning of the storage period, the hydrocolloids perform their role and maintain the complete system thicken and stable, while over time these bonds and interconnection become relaxed and baking stability changes together with an increased viscosity.

Texture properties

Texture properties of processed cheeses are mostly determined by their composition especially fat and proteins, structural arrangements of present components (e.g. microstructure) and physicochemical state of the constituents (O'Callaghan and Guinee, 2004). The importance of textural properties depends on the final usage of processed cheeses. In this study firmness and stickiness were determined. Firmness is maximum force obtained during penetration test, while stickiness is measured as a force necessary to remove probe. Firmer and stickier samples are usually more difficult to

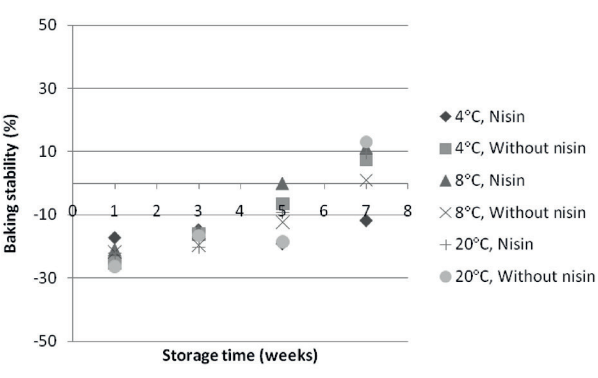

Figure 1. Baking stability (%) in different processed cheese samples over time. Values represent mean±standard deviation (n=3)

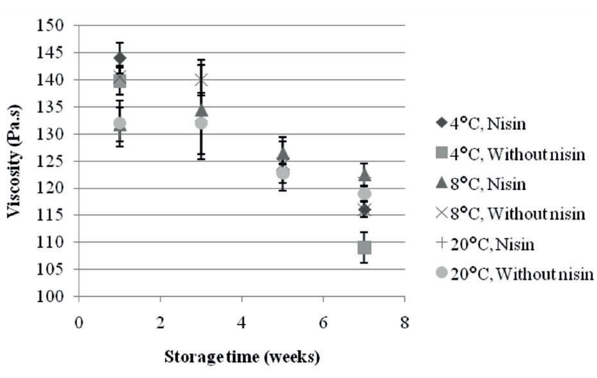

Figure 2. Viscosity values in different processed cheese samples over time. Values represent mean±standard deviation (n=3)

spread. Changes of textural properties of processed cheese samples are shown on Figure 3. The significant changes of measured parameters during the experimental period were not found. Also, no significant effects of storage temperature or nisin were found. Texture properties of processed cheeses such as hardness and adhesiveness are dependent on protein concentration (Sołowiej et al., 2014) and the type of fat (Cunha et al., 2010). Literature data on texture changes of processed cheeses are very rare. However, Schar and Bosset (2002) consider that texture changes of processed cheeses could be due to the loss of water vapour, hydrolysis of polyphosphates, changes in ionic equilibrium, formation of crystals, reactions induced by heat-stable enzymes and storage temperature. No changes found in our study indicated that processed cheeses are very stable product, regarding texture and independent on the storage temperature.

Colour change

Calculated values of ΔE, WI and YI are presented in Table 3. The results showed significant colour changes between samples kept at different temperatures at a given sampling day. Additionally, during the observed period, colour changed in the same samples. Samples became whiter and less yellow, compared to freshly prepared samples. When analysing block processed cheese during 3 months of storage, Awad et al. (2004) found that the cheese samples became darker. The changes were even more pronounced for samples stored at room temperature compared to low storage temperature (7 °C). Therefore, it is to be expected that colour of processed cheese samples will slightly change over time, due to chemical reaction and changes in composition of samples.

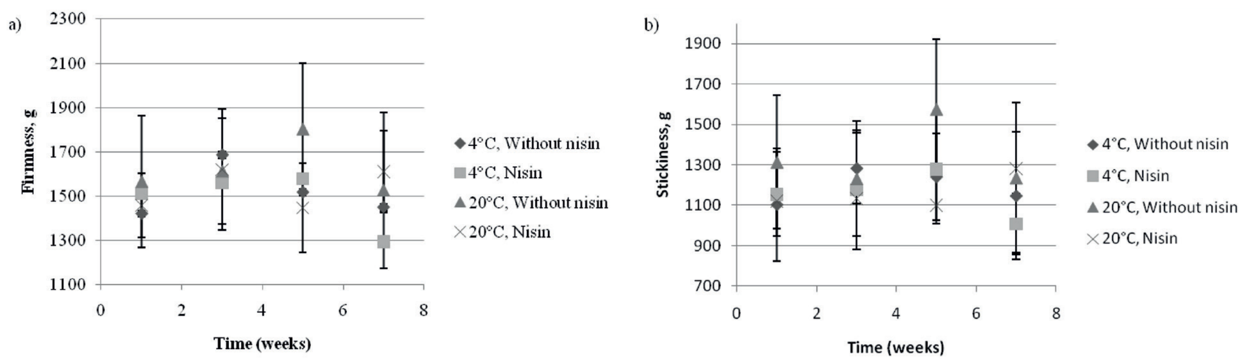

Figure 3. Values for firmness (a) and stickiness (b) in different samples over time. Values represent mean±standard deviation (n=3).

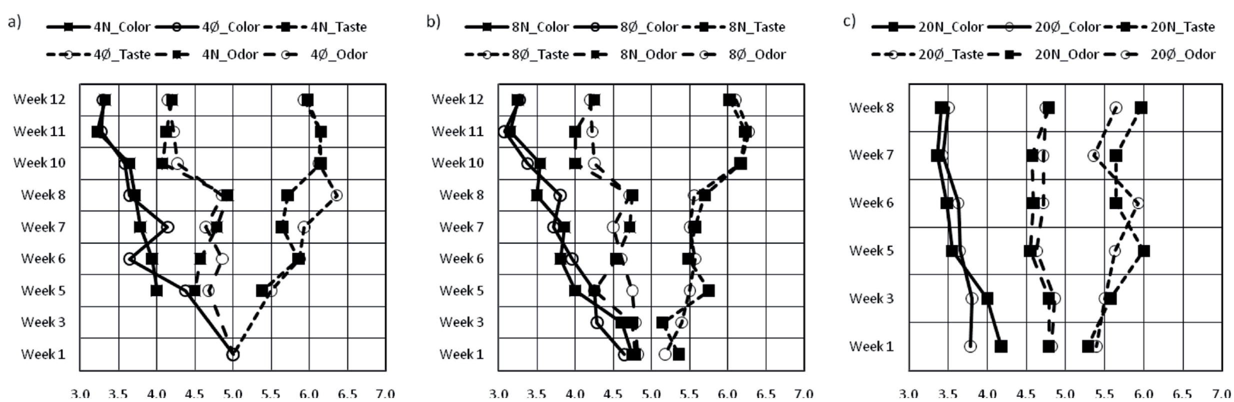

Figure 4. The deviation of sensorial attributes (colour, odour and taste) from the "control" sample during storage at different temperatures. "Control" sample present freshly prepared sample without added nisin and stored at 4 °C during 1 week. a) Storage temperature 4 °C; b) Storage temperature 8 °C; c) Storage temperature 20 °C. Legend: 4Ø - 4 °C, without nisin; 8Ø - 8 °C, without nisin; 20Ø – 20 °C, without nisin; 4N - 4 °C, with nisin; 8N - 4°C, with nisin; 20N - 4 °C, with nisin

These instrumental measurements of colour changes were confirmed by sensory evaluation of processed cheese samples. This is in line with definition given by Mokrzycki and Tatol (2011), who explained that one can determine an important and noticeable changes in colour only for ΔE values ≥ 2 (0 < ΔE < 2 - only experienced observers may notice the difference; 2 < ΔE < 3.5 - average observers may notice the difference; and ΔE > 3.5 - clear difference in colour is noticed).

Sensory analysis

The results obtained during sensorial analysis are presented in Figure 4. It can be seen that the two mostly affected attributes of processed cheese were colour and taste. At all times, and more pronounced starting from week 5, these parameters deviated in greater extend from the "control" sample, which was presented as value 5 or central line (Figure 4). It is of note that colour deviated in the direction of "less intense than control samples" (score < 4), with the most often defects being "change of colour" and "surface colour inequality". On the other side, the taste deviated in the direction of "more intense than control sample" (score > 6), with the most frequent defects being "unpleasant aftertaste" and "starchy taste". It is of note that the longer the storage period, the more deviation of colour and taste from the control samples could be observed. This is in line with CVS measurements of colour (Table 4). The observed sensorial changes most probably occurred due to the chemical changes rather than microbiological changes in analysed samples. Similarly to these obtained results, Awad et al. (2004) reported that all cheese samples exhibited slight changes in the sensorial acceptability values over the storage period of 3 months. Results obtained in this study indicated that the addition of nisin had no influence on the sensorial attributes of samples (Figure 4), as nisin does not cause intensive alteration in sensory properties of food products. Likewise, when nisin A was added to high-fat chilled dairy dessert, a milk-based pudding, no significant difference was observed between samples with and without nisin (Oshima et al., 2014).

Table 3. Colour (total colour difference, ΔE, whitening index, WI and yellowness index, YI) determined in different processed cheese samples stored at different temperature (4 °C, 8 °C and 20 °C) and prepared without and with nisin (12.5 ppm). Values represent mean ± standard deviation (n=3)

Total color difference (ΔE), mean±standard deviation (n=3)						
	Storage temperature, 4 °C		Storage temperature, 8 °C		Storage temperature, 20 °C	
Time (weeks)	With nisin	Without nisin	With nisin	Without nisin	With nisin	Without nisin
1	1.41±0.15^{aBC}	0.64±0.32^{aA}	1.70±0.35^{aC}	1.20±0.53^{aB}	2.28±0.11^{aD}	1.03±0.37^{aAB}
3	2.37±0.56^{bB}	1.11±0.22^{bA}	2.38±0.50^{bB}	2.03±0.97^{bB}	3.57±0.57^{bC}	1.79±0.46^{bAB}
5	1.57±0.50^{aA}	1.87±0.57^{cA}	3.76±0.59^{cC}	2.90±0.46^{cB}	3.75±0.36^{bC}	2.78±0.57^{cB}
7	3.47±0.74^{cA}	4.12±0.24^{dB}	3.83±0.46^{cAB}	3.42±0.25^{cA}	4.34±0.39^{cB}	3.87±0.45^{dAB}
Time (weeks)	Whitening index (WI), mean±standard deviation (n=3)					
1	78.20±0.36^{aAB}	78.99±0.43^{aC}	79.33±0.56^{aCD}	79.77±0.42^{aD}	77.86±0.49^{aA}	78.87±0.67^{aBC}
3	80.65±0.38^{bAB}	79.64±0.35^{bA}	81.02±0.57^{bBC}	79.88±1.68^{aA}	81.84±0.71^{cC}	80.41±0.52^{bAB}
5	79.06 ±0.81^{cA}	80.30±0.57^{cBC}	81.39±0.63^{bcD}	81.04±0.58^{bCD}	79.95±0.70^{bB}	79.92±0.57^{bB}
7	81.53±0.47^{dA}	82.74±0.30^{dC}	81.92±0.53^{cAB}	82.10±0.34^{bB}	82.20±0.28^{cB}	82.24±0.57^{cBC}
Time (weeks)	Yellowness index (YI), mean±standard deviation (n=3)					
1	22.33±0.11^{aC}	20.29±0.67^{aA}	21.7±0.76^{aBC}	20.29±0.13^{aA}	23.95±0.16^{aC}	21.35±20.41^{aB}
3	20.33±0.63^{cB}	19.35±0.83^{bAB}	19.22±0.96^{bAB}	19.53±1.15^{aAB}	19.27±0.94^{cAB}	18.92±20.65^{bA}
5	21.11±1.00^{bBC}	19.44±0.73^{abA}	20.17±0.72^{bAB}	19.53±0.90^{aA}	22.13±0.83^{bC}	21.14±19.93^{aBC}
7	19.86±0.12^{cC}	17.79±0.67^{cA}	19.51±0.72^{bC}	17.47±0.88^{bA}	19.67±0.09^{cC}	18.68±18.27^{bB}

Means in the same column with different small letters and means in the same row with different capital letters are significantly different ($p<0.05$)

Conclusion

Microbiological results indicated that processed cheese samples were rather stable over the experimental period. The most effective combination in controlling ACC was the addition of nisin (12.5 ppm) and low storage temperature (4 °C). Nevertheless, no change / increase was observed for PCC or YM. Regarding quality characteristics of processed cheese samples, textural changes were less prominent in the samples. Significant changes were determined for parameters such as colour, viscosity and baking stability. It can be assumed that these changes occurred as a result of chemical changes, rather than microbiological. These results were also confirmed by sensory analysis. Since these quality parameters are mainly consumer oriented, meaning that consumers may complain on changes in the sensory profile, colour and viscosity, technological modifications are needed to optimize these parameters, bearing in mind that the safety of the product is not affected. Future research should focus on optimizing the duration of shelf life, and improving the sensory profile.

Učinak nizina i temperature skladištenja na parametre kvalitete topljenog sira

Sažetak

Cilj ovog rada bio je procijeniti učinak nizina i temperature skladištenja na mikrobiološka, fizikalno-kemijska i senzorska svojstva topljenog sira. Uzorci topljenog sira pripremljeni su bez i sa 12,5 ppm nizina, pohranjeni na različitim temperaturama (4 °C, 8 °C i 20 °C). U uzorcima topljenih sireva pripremljenim bez nizina, broj aerobnih kolonija (ACC) značajno je porastao tijekom eksperimentalnog razdoblja, dok u uzorcima s nizinom nije utvrđen značajniji porast. Pri nižoj temperaturi skladištenja (4 °C) učinkovitost nizina bila je izraženija. Kvasci i plijesni (YM) i broj psihrotrofnih kolonija (PCC) bili su ispod granice kvantifikacije tijekom cijelog eksperimentalnog razdoblja. Također, nisu utvrđene značajne promjene pH vrijednosti i teksturalnih svojstava (čvrstoća i ljepljivosti) uzoraka. Senzorska analiza potvrdila je da su uzorci topljenog sira postali bjelji i manje žuti, u usporedbi sa svježim uzorcima. Viskoznost svih uzoraka smanjena je tijekom eksperimentalnog razdoblja, što je bilo u skladu s rezultatima stabilnosti pečenja. Dobiveni rezultati potvrdili su potencijal korištenja ove vrste topljenog sira u pekarskoj industriji.

Ključne riječi: topljeni sir, parametri kvalitete, nizin, temperatura skladištenja

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