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# The impact of different starter cultures on fat content, pH and SH dynamics in white brined cheese production

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**Summary**.White brined cheese is a specific dairy product for Balkan Peninsula countries, Mediterranean, North Africa, Eastern Europe and some parts of Asia. The survey was conducted in 2016 at a dairy industry laboratory in R. of Macedonia. In this research work the influence of three different starter cultures of three white brined cheese variants (A, B, C) has been examined regarding the fat content dynamics. The starter culture in variant A (SMCH-5) contained following bacteria strains: Lb. bulgaricus, Str. thermophilus and Lb. acidophilus. In the variant B (Choozit Feta A) the follow bacteria strains were included: Lac. lactis ssp. lactis, Lac. lactis ssp. cremoris, Str. thermophilus, Lb. bulgaricus and Lb. helveticus. The variant C (MOTC 092 EE) was a combination of the strains: Lac. lactis ssp. lactis, Str. thermophilus, Lb. bulgaricus, Lb. helveticus and Lb. casei. The impact of the above mentioned three different starter cultures was determined over the fat content, pH and SH during the process of ripening of the white brined cheese.

**Keywords**:white-brined cheese, starter cultures, fat content, pH, SH, dynamic

## Introduction

Cheese is the generic name for a group of fermented milk-based food products produced worldwide in a wide range of flavours and forms. Although there is no reliable estimate of the number of cheese varieties produced throughout the world, well over 1000 named varieties are recognized. Cheese is the quintessential convenience food; indeed, it has been said that there is a cheese for every taste preference and a taste preference for every cheese. (Eck, A. and Gilles J. – C., 2000)

Cheese manufacture, which dates from 6000–7000 BC, is essentially a method for preserving the nutritive value of milk through fermentation, removal of moisture and addition of salt.

It is fascinating that such a wide range of flavours can be produced from the basic ingredients of cheese which are milk, starter cultures, salt and often rennet.

Cheese is a fermented food product in which lactose is converted to lactic acid by the action of bacteria. The strains of lactic acid bacteria used to acidify milk are usually carefully selected and deliberately added to milk as a culture known as a starter.

White brined cheese has a great tradition in R. Macedonia and is usually produced from cow milk. The increased consumption of white brined cheese contributes to the necessity to be produced in almost all dairy facilities in industrial way: milk pasteurization, adding ingredients (calcium chloride, color, rennent.), and also the addition of starter cultures for continuous milk acid fermentation.

According to El Soda at al. (2003), the use of commercial starter cultures in an industrial way of cheese production is necessary for obtaining a final product with a standard identifiable feature. A variety of bacteria cultures are available for making specific cheese types. These bacteria, commonly called starters, are added to the milk after pasteurization and at specific temperatures. They are allowed to work for specific time periods depending on the type of cheese. During this period the bacteria consumes the lactose which is milk sugar.

The most important function of the starter cultures is the production of lactic acid and the release of enzymes during the fermentation process of white brined cheese (Leroy and de Vuyst, 2004).

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## Material and methods

As for this research the white brined cheese was manufactured from pasteurized cow milk in a local dairy plant "Milkom" – v. Nogaevci, Gradsko, R. Macedonia. Regarding the production the raw milk was supplied from Gradsko region in Macedonia. The chemical composition of the milk used for the manufacturing of white cheese was 12.13% total solids, 3.7% fat, 3.21% protein, 0.67% ash, and 4.55% lactose. The pH of the milk was 6.49 and it was pasteurized at 72 °C for 15 seconds and cooled at 34 °C. The curding was done at temperature of $34^{o}$ C. First the following starter cultures were added: for white brined-cheeseVariant A – SMCH-5, for white brined cheeseVariant B – Choozit Feta A and for white brined cheese Variant C – MOTC 092 EE. Then the $CaCl_2$ 0.02% and blego color 10 ml/100 l milk was added. The cow milk was coagulated with chymosin rennet (Chymax Extra Powder 1.5 g/100 l milk) completed in 45 min. Further on, the curd was cut in cubes of 1 cm3, resting for 5 minutes and afterwards pressed in cheese mold for 3 hours. Cheese blocks were placed in tinned cans filled with brine solution of 8g NaCl/100g. During the ripening period of 30 days the cheese was held at 15–17 °C, and then kept at 2–4 °C. (Figure 1 White-brined cheese technology).

In order to determine their influence on fat content, pH and SH in white brined cheese three different types of starter cultures were used. There were three variants of cheese produced which differ by starter cultures used in production process (Var. A – SMCH-5, product by LB Lactis – Bulgaria, Var. B – Choozit Feta A – product by Danisco – Denmark and Var. C MOTC 092 EE (produced by Sacco Clerici).

The determination of the content of milk fat in cheese is determined by the Gerber method (Caric at al. 2002). The pH of milk and cheese samples was measured using a digital pH meter (digital pH meter, model MP120FK Mettler Toledo, Greifensee, Switzerland). SH was measured according to Soxlet Henkel method

The determination of milk fat content, pH and SH of white brined cheese was examined on the 1st day, 10th day, 30th day and the 60th day. Further on, the standard statistical method (Najchevska, 2002) was used for statistical presentation of the analyzed data as well as the F-test for analysis of the variance in tested cheese variants.

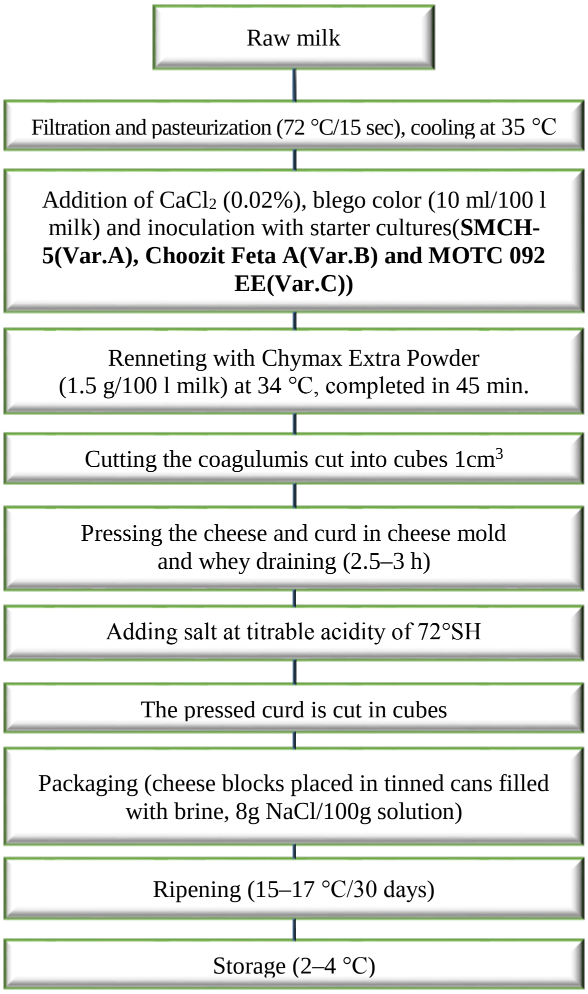


Figure 1. Modified protocol of white cheese production (Ozcan at al.2012)

## Results and discussion

Milk fat remains incorporated between the protein matrix and the curd, although a very small part is eliminated through the whey after cutting and pressing the curd.

Milk fats influence rheological and sensory characteristics of white brined cheese. The greater amount of milk fat helps generate white brined cheese with softer consistency and better sensory properties, unlike the low fat milk percentage which helps generate white brined cheese with hard consistency and its sensory properties are less expressed.

The fat content regarding the examined three varieties of white brined cheese is presented in Table 1, and in Figure 1. According to the results depicted in Tables 1 it can be concluded that fat content in the white brined cheese (var. A, var. B and var. C) at the first day of production was the following: 20.96% (var. A); 21.36% (var. B)и 21,32% (var. C).

Significant changes regarding the further period of ripening have not been registered which is shown in the graph bellow.

Continuous increase of fat content was noticed to all examined variants of white brined cheese (var. A var. B and var. C) without significant peaks. After 60 days of ripening period, Variant A had shown 22.40% of fat content, Variant B had shown 22.42% of fat content and Variant C had shown 22.34% of fat content.

Table 1.

Fat content dynamics in white brined cheese

| Index | Variant A (SMCH-5) | | | | Variant B (Choozit Feta A) | | | | Variant C (MOTC 092 EE) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day |
| x | 20.96 | 21.48 | 22.02 | 22.40 | 21.36 | 21.84 | 22 | 22.42 | 21.32 | 21.8 | 21.94 | 22.34 |
| Min | 20.60 | 21 | 21.70 | 22.10 | 21.20 | 21.60 | 21.80 | 22.20 | 21 | 21.5 | 21.6 | 22.2 |
| Max | 21.20 | 21.70 | 22.30 | 22.60 | 21.60 | 22.10 | 22.20 | 22.60 | 21.5 | 22 | 22.2 | 22.5 |
| Sd | 0.230 | 0.295 | 0.239 | 0.187 | 0.167 | 0.207 | 0.187 | 0.148 | 0.217 | 0.212 | 0.23 | 0.152 |
| Cv | 1.098 | 1.373 | 1.084 | 0.835 | 0.783 | 0.949 | 0.850 | 0.662 | 1.017 | 0.973 | 1.049 | 0.679 |

The presented data are in correlation with the conclusions of Dozet et al. (1996) emphasizing that in the production process of white brined cheese, lipolytic reactions are not significantly expressed which however differs for blue cheeses, where more lipolytic processes and more changes in fat content percentage are noticed.

Popović, Vranješ et al. (2011) had similar results in their study regarding the fat content in Sjenichko cheese. They have determined that the average of fat content was 22.97% (minimum 21.97% and maximum 23.97% of fat content) which appears as similar to the results presented in this paper.

The results presented in this survey are quite similar to the results provided by Chomakov at al. (2000). Their values for fat content in white brined cheese are in the range limits of 21–25%.

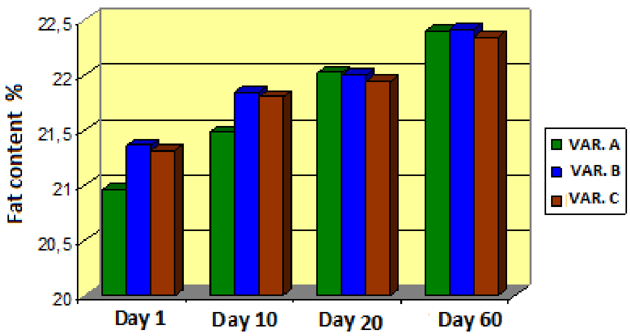


Figure 2. Fat content dynamics in white brined cheese

The increase of fat content in white brined cheese from this survey is also followed by increase of the dry fat matter content. The energy value of cheese mostly depends on the fat content. It is of a great importance that the milk during the industrial production should not be transported in long lines, causing increased fat loss, which in total will effect on product amount.

With the use of Fisher's-test statistically significant differences among the tested varieties were not determined, so it can be concluded that the starter cultures do not affect the fat content in white brined cheese (table 2).

Table 2

Analysis of variance for the fat content in white brined cheese

| Source of variation | Sum of squares (SQ) | Degree of freedom (DF) | Variance | F-value |
|---|---|---|---|---|
| Total | 0.3374 | 14 | | |
| Between groups | 0.0174 | 2 | 0.0087 | ns 0.32 |
| In groups | 0.32 | 12 | 0.0266 | |

ns – not statistically significant; • significance level $p < 0.05$; ** significance level $p < 0.01$ $F_{0.05} = 3,74$ $p < 0,05$; $F_{0.01} = 6.51$ $p < 0.01$

Active acidity (pH) is defined as concentration of hydrogen ions. This parameter according Baltadzhieva (1993) has a control function and reflects the buffering capacity of the cheese. By increasing the concentration of hydrogen or hydroxyl ions to some extent, active acidity is not changed. The pH value of the cheese is most affected by phosphates, carbonates, citrates, casein, albumin and globulins. The dynamics of the active acidity in tested variants of cheese is shown in Tables 5, 6 and 7, and also in Figure 2.

In this survey the pH values at the first day after production of the three variants of cow's brined cheeses were as followed: 4.93 (Var. A); 4.95 (Var. C) and 5.01 (Var. B). The content of starter cultures is so important for the dynamics of the active acidity.

According to Gruev (1995) the most important bacteria for fermentation process in brined cheese production is Str. Lactis. This bacterium has role to make hydrolytic degradation of cheese paracasein. In further stages of ripening particularly important is bacterium Lac. Casei. While yogurt culture composed of Str. thermophilus and Lac. bulgaricus practically does not participate in the maturation process because they develop at temperatures above 15 °C and salt concentration between 6 to 8%.

The activity of starter cultures and the decrease of pH is expressed on the 10th day when the pH was 4.66 (Var. A); 4.71 (Var. B) and 4.69 (Var. C). After the 10th day came to accumulation of lactic acid with the transformation of lactose which has negative affects on the bacteria. That's the reason for decreasing the process of acidification and autolysis on lactic acid bacteria cells.

The value of active acidity after period of one month fermentation was similar for each variant white brined cheese: 4.5 (Var. A); 4.54 (Var. B) and 4.52 (Var. C).

Table 3.

Dynamics of active acidity (pH) of white brined cheese

| Index | Variant A (SMCH-5) | | | | Variant B (Choozit Feta A) | | | | Variant C (MOTC 092 EE) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day |
| x | 4.93 | 4.66 | 4.5 | 4.42 | 5.01 | 4.71 | 4.54 | 4.48 | 4.95 | 4.69 | 4.52 | 4.45 |
| Min | 4.87 | 4.61 | 4.44 | 4.4 | 4.91 | 4.68 | 4.48 | 4.43 | 4.88 | 4.6 | 4.43 | 4.41 |
| Max | 5.02 | 4.71 | 4.55 | 4.45 | 5.09 | 4.75 | 4.61 | 4.52 | 5.04 | 4.75 | 4.6 | 4.5 |
| Sd | 0.058 | 0.045 | 0.051 | 0.019 | 0.082 | 0.032 | 0.049 | 0.042 | 0.06 | 0.06 | 0.067 | 0.036 |
| Cv | 1.174 | 0.972 | 1.133 | 0.423 | 1.628 | 0.688 | 1.09 | 0.934 | 1.207 | 1.275 | 1.482 | 0.805 |

The further reduction of active acidity level was with lower dynamics. At 60-th day the lowest pH value was determined in variant A (4.42), then the variant C (4.45), while the highest pH was measured in variant B (4.48).

These changes are regarded identical to the research of Radulović et al. (2011) who suggested that lactococcus achieve their maximum untill the 15th day of ripening, but they were not entirely extinct in the following period. High level of this bacteria 106 cfu g[1] is registered at the 60th day of ripening.

Slightly higher results from this survey were presented Smiljanić et al. (2014). In their studies the value of pH after 60 days fermentation was 4.6, which can be explained by the fact that in traditional manufacturing process of white brined cheese starter culture is not added like in our case.

The obtained data for pH of white brined cheese in this survey are in correlation with the results by Chobanova Vasilevska (2007) where were noticed variations of pH from 4.50 to 4.8, and also by Ostojić and Mesner (1978) where was determined an average of pH 4.44.

According to Kostova (2013) the pH of traditional Macedonian white brined cheese is in range between 4.04 to 5.05.

Similar results to ours were also presented by Talevski (2011). In his survey he produced three variants of white brined cheese with three different starter cultures. The pH of white brined cheeses were between 4.54 to 4.62.

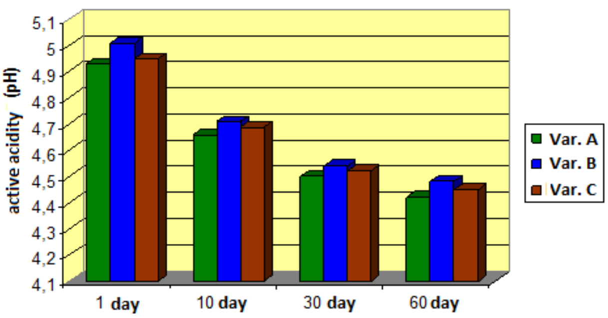


Figure 3. Dynamics of the active acidity (pH) in white brined cheese

Table 4.

Analysis of variance for the active acidity (pH) in white brined cheese

| Source of variation | Sum of squares (SQ) | Degree of freedom (DF) | Variance | F-value |
|---|---|---|---|---|
| Total | 0.0231 | 14 | | *4.25 |
| Between groups | 0.00958 | 2 | 0.00479 | |
| In groups | 0.01352 | 12 | 0.001126 | |

ns – not statistically significant; • significance level $p < 0.05$; ** significance level $p < 0.01$ $F_{0.05} = 3.74$ за $p < 0.05$; $F_{0.01} = 6.51$ за $p < 0.01$

Starter cultures have minimal influence on pH value of white brined cheese at level $p < 0.05$ which can be noticed with positive Fisher test (table 4).

The dynamics of the titratable acidity of the three varieties of white brined cheese is shown in table 5, and also in and figure 4. The starting values in tested variants were: 51.20°SH (var. A); 48.40°SH (var. B) 50.40°SH (var. C). These results for titratable acidity were quite similar with results presented by Tratnik at al. (2000) for feta cheese obtained from full fat and partly skimmed milk. Titratable acidity between 47.27 and 61.81°SH.

On the tenth day of production were noticed a continuous increase of titratable acidity in all variants. The minimum value at that point of measurement was established in white brined cheese (variant B) produced from Choozit Feta A (69.20°SH), while the maximum reached the white brined cheese (variant A) produced from SMCH-5 (72°SH).

After 30 days fermentation period, the titratable acidity values of three variants white brined cheese were as follows: 79.20°SH (Variant A); 76.40°SH (Variant B) and 77.60°SH (Variant C).

In that stage of fermentation, the increase of titratable acidity is mostly affected by Lactobacillus which are tolerant to low pH and high salt concentration. According to Núñez (1978), at the initial stage prevailing streptococci, but due to the high salt concentration and the inhibitory action of the active acidity comes to their extinction and its place is taken by Lactobacillus.

These results approximating the optimal acidity in this period noted by Stojiljkovic (2007) indicating that the cheese titratable acidity in 20–25 days should be 62–74°SH. Higher titratable acidity causes friable and sour cheese, while the lower acidity has the opportunity to get to faster deterioration due to lack of lactic acid.

Table 5.

Dynamics of titratable acidity (°SH) of titratable acidity in white brined cheese

| Index | Variant A (SMCH-5) | | | | Variant B (Choozit Feta A) | | | | Variant C (MOTC 092 EE) | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day | 1 day | 10 day | 30 day | 60 day |
| x | 51.20 | 72 | 79.20 | 86.40 | 48.40 | 69.20 | 76.40 | 82 | 50.40 | 70 | 77.60 | 84.40 |
| Min | 48 | 66 | 76 | 84 | 46 | 64 | 74 | 80 | 48 | 68 | 74 | 82 |
| Max | 54 | 74 | 82 | 88 | 52 | 74 | 78 | 84 | 52 | 74 | 82 | 86 |
| Sd | 2.280 | 3.464 | 2.280 | 1.673 | 2.191 | 3.633 | 1.673 | 1.414 | 2.191 | 2.449 | 3.578 | 1.673 |
| Cv | 4.454 | 4.811 | 2.879 | 1.937 | 4.527 | 5.250 | 2.190 | 1.725 | 4.347 | 3.499 | 4.610 | 1.983 |

Generally, it can be concluded that the main lactic acid fermentation process for white brined cheese is completed within 30 days and after that period the intensity of lactic acid fermentation process is getting lower. Rapid production of lactic acid and the increase of titratable acidity helps to create a greater amount of monocalcium para-casein which cause better gluing of curd grains and compact cheese pieces.

After 30 days fermentation period, the titratable acidity values of three variants white brined cheese were as follows: Variant A 86.40°SH, Variant B – 82°SH and Variant C – 84.40°SH.

According to Kostova (2013), the titrable acidity in white brined cheese is between 80°SH and 96°SH. Our results correspond with the results of Sjenichkoto cheese produced from sheep's milk where titratable acidity had extremes of 57.54 to 97.73°SH. (Ružić Muslićи cop., 2011).

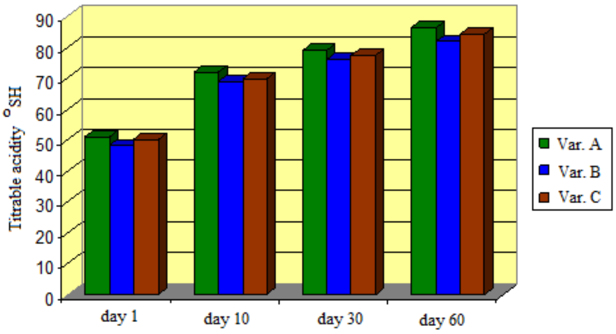


Figure 4. Dynamics of titrable acidity (°SH) in white brined cheese

At the table above are presented the results of analysis of variance for titratable acidity in three variants of white brined cheese. From the obtained results it is noticed that the starter cultures have significant impact at level $p < 0.01$ and have influence on white brined cheese titrable acidity. Obtained F-value (9.58) was greater than the tabular values of both levels.

Table 6.
Analysis of variance of titratable acidity (°SH) in white brined cheese

| Source of variation | Sum of squares (SQ) | Degree of freedom (DF) | Variance | F-value |
|---|---|---|---|---|
| Total | 78.934 | 14 | | **9.58 |
| Between groups | 48.534 | 2 | 24.267 | |
| In groups | 30.40 | 12 | 2.533 | |

ns – not statistically significant; • significance level $p < 0.05$; ** significance level $p < 0.01$ $F_{0.05} = 3.74$ $p < 0.05$; $F_{0.01} = 6.51$ $p < 0.01$

## Conclusion

Significant differences in the content of milk fat during the fermentation process among all three white brined cheese variants (A, B and C) produced by different types of starter cultures have not been found. According to the analyzed data it can be confirmed and concluded that starter cultures do not influence the content of milk fat during the process of fermentation of white brined cheese.

The content of milk fat was presented such as: 22.34% (variant C), 22.4% (Variant A) to 22.42% (variant B). The starter cultures that were used in the production process of the three different variants (A, B and C) of white brined cheese had a minimal impact on the dynamics of the active acidity The pH values of all three variants of cheese (A, B and C) after two months of ripening period were as follow: 4.42 (Var. A), 4.48 (Var. B) and 4.45 (Var. C). The starter cultures that were used in the production process of the three different variants (A, B and C) of white brined cheese had a minimal impact on the dynamics of the titrable acidity. The SH values of all three variants of cheese (A, B and C) after two months of ripening period were as follow: 86.40 (Var. A), 82 (Var. B) and 84.40 (Var. C).

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## CONTRIBUTION

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## CONFLICT OF INTEREST

The authors declare no conflict of interest.

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