ANNALS OF THE UNIVERSITY OF CRAIOVA

Series: ✓ *Biology*
✓ *Horticulture*
✓ *Food products processing technology*
✓ *Environmental engineering*

Vol. XXX (LXVI) - 2025

EFFECT OF STARTER CULTURE CONCENTRATION ON THE PHYSICOCHEMICAL PROPERTIES OF NATURAL YOGURT

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Keywords: n*atural yogurt, starter cultures, yogurt quality, physicochemical properties*

ABSTRACT

The study analyzes the influence of different concentrations of mixed starter cultures of Lactobacillus delbrueckii subsp. bulgaricus and Streptococcus thermophilus on the physicochemical characteristics of natural yogurt. Yogurt was prepared from milk with 3.5 % fat, to which starter cultures were added at proportions of 0.5 %, 1 %, and 2 % of the milk volume for three separate samples. After homogenization and incubation, the obtained samples were analyzed from a physicochemical point of view. The different concentrations of the starter culture have a direct impact on the quality and physicochemical properties of the yogurt, affecting parameters such as acidity, texture, syneresis, and flavor. The determinations carried out included titratable acidity, fat content, moisture, and pH. The results obtained highlight how the level of inoculation with starter cultures influences the final quality of natural yogurt.

INTRODUCTION

Starter cultures are one of the most important factors in providing the technological and organoleptic characteristics of fermented dairy products (Amani et al., 2017). *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*, used in a mixed culture, have a symbiotic relationship in milk through the exchange of metabolites necessary for the growth of each bacterium (Arioli et al., 2017; Dan et al., 2019). The production of metabolites such as amino acids and short peptides contributes to the formation of aromatic substances in yogurt, giving it its characteristic flavor (Dan et al., 2019; Carroll et al., 2016; Cheng, 2010). Yogurt occupies an important place in a balanced diet due to its high nutritional value. It is an excellent source of complete proteins, vitamins (especially vitamins B2 and B12), essential minerals (such as calcium and phosphorus), and probiotic bacteria, which help maintain the health of the digestive system (Sumi et al., 2023). Being one of the most representative probiotics, yogurt has been reported to influence the composition of the intestinal flora by reducing pathogenic bacteria (Sumi et al., 2023; Aslam et al., 2020).

Regular consumption of natural yogurt brings multiple health benefits. The probiotics in yogurt, such as *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, help balance the intestinal flora, contributing to healthy digestion (Hadjimbei et al., 2022).

Due to its rich calcium and phosphorus content, yogurt helps maintain bone health and prevent conditions such as osteoporosis (Hadjimbei et al., 2022; Bibi et al., 2021). Nutrients, especially vitamin B12 and probiotics, support the proper functioning of the immune system. Compared to milk, yogurt is easier to tolerate by people with lactose intolerance, as the bacteria present in the product help break down lactose (Khiaosa-Ard et al., 2022). In addition to these benefits, natural yogurt has a low calorie and fat content, making it an ideal choice for people who want to maintain a healthy weight or follow a balanced diet (Sumi et al., 2023; Sigala-Robles et al., 2022).

Regular consumption of natural yogurt brings multiple health benefits, such as intestinal regulation, immunomodulatory action, prevention of diabetes, and prevention of heart diseases (Sumi et al., 2023; Hadjimbei et al., 2022). Yogurt is a good source of calcium because its absorption is facilitated by galacto-oligosaccharides and casein phosphopeptides in dairy products (Sumi et al., 2023; Hadjimbei et al., 2022).

Yogurt, like milk, also contains high levels of short-chain fatty acids (Sumi et al., 2023; Hadjimbei et al., 2022) and various peptides derived from milk proteins, and it has been suggested that these components of yogurt help reduce intestinal inflammation and protect the immune system (Sumi et al., 2023; Khiaosa-Ard et al., 2022). In addition, a large cohort study from 21 countries reported that the consumption of full-fat dairy products, including yogurt, reduces the risks of metabolic syndrome, high blood pressure, and diabetes (Sigala-Robles et al., 2022).

MATERIAL AND METHODS

For the preparation of natural yogurt, the following primary and auxiliary materials were used: pasteurized whole cow’s milk with 3.5 % fat content and a defined lyophilized starter culture. Three experimental trials were conducted using different concentrations of lactic cultures (0.5 %, 1 %, and 2 % of the milk volume). For the preparation of four portions of yogurt, the following ingredients were required: 1000 mL of pasteurized cow’s milk and 10 g of lyophilized starter culture. The milk was heated to the inoculation temperature of 40–45 °C using a water bath, and the temperature was continuously monitored.

Starter cultures were added at the specified concentrations for each trial, and the mixture was thoroughly homogenized to ensure even distribution of bacterial cells.The inoculated milk was poured into pre-sterilized containers and incubated in a thermostat-controlled incubator at 44 °C for 2.5–3 hours, maintaining a constant temperature throughout the process. The acidification process was considered complete when the pH reached 5.5. After incubation, the yogurt samples were pre-cooled at 20 °C for 3–4 hours to stop further fermentation. The product was then stored at 4 °C for 4–8 hours before sensory evaluation or consumption. The shelf life of unopened yogurt was 7–14 days under refrigeration, while after opening, consumption within 7 days was recommended. Physicochemical analyses were performed on all yogurt samples to evaluate the impact of starter culture concentration on product quality. Determinations included titratable acidity, fat content, moisture content, and pH. Each sample was analyzed individually, and all measurements were conducted in triplicate to ensure accuracy and reproducibility. Data were recorded and tabulated for subsequent comparison among the three different starter culture concentrations.

RESULTS AND DISCUSSIONS

Three natural yogurt samples were prepared using different concentrations of starter culture: 0.5 %, 1 %, and 2 %, and each sample was analyzed individually for physicochemical properties. The evaluated characteristics included titratable acidity, pH, and fat content, to confirm the presence of lactic fermentation, the active influence of the starter cultures, and the classification of the final product as yogurt. Measurements were performed for the three samples at different time intervals. The first measurements were taken immediately after inoculation, at the end of the yogurt production process. Subsequent measurements were taken 3 hours after production, and the final measurements 6 hours after production. The results obtained in the study are illustrated in the following figures, which highlight the main differences and trends observed.

Figure 1 are illustrated the titratable acidity (°T) values of the three yogurt samples, information that contributes to the comparison and interpretation of their characteristics.

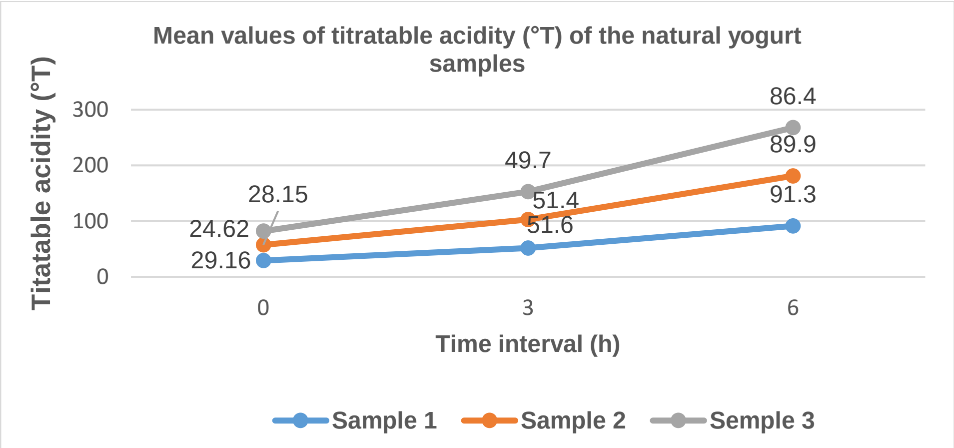

Figure 1. Mean values of titratable acidity (°T) of natural yogurt samples

The titratable acidity of the samples continued to increase during fermentation, with values ranging from 24.62, very low at the time of inoculation, up to 91.3 at the moment fermentation was stopped by cooling.

The analyses performed fully comply with the SR ISO 11869:1997 standards, ensuring that all measurements were conducted according to internationally recognized procedures.

Figure 2 shows the mean pH values of the yogurt samples, allowing for a comparison of the acidity levels between the different samples.

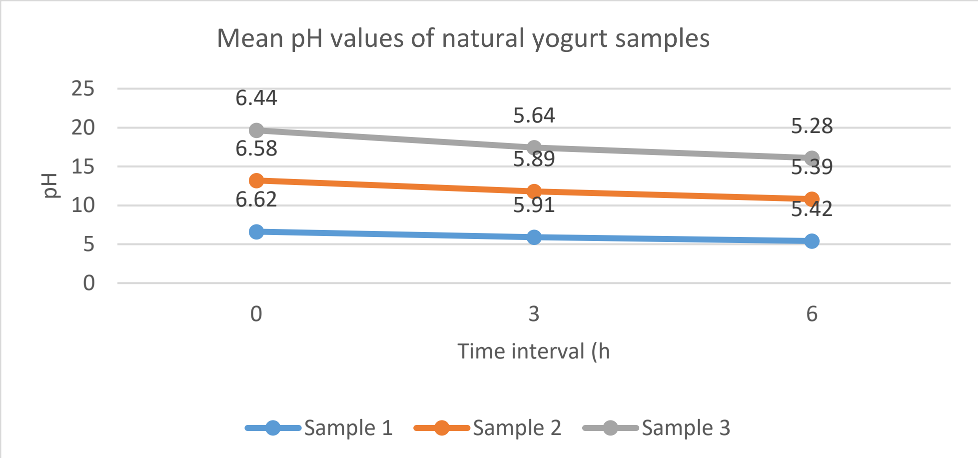

Figure 2. Mean pH values of natural yogurt samples

The pH values of the analyzed samples ranged from slightly basic immediately after inoculation to mildly acidic at the end of yogurt cooling, and acidic a few hours after completing the yogurt production stages. The most basic samples had pH values between 6.62 and 6.44, and at the final measurement, the pH stabilized between 5.28 and 5.42.

Figure 3 shows the average fat content (%) of the natural yogurt samples, allowing for an easy comparison of the values between the different samples.

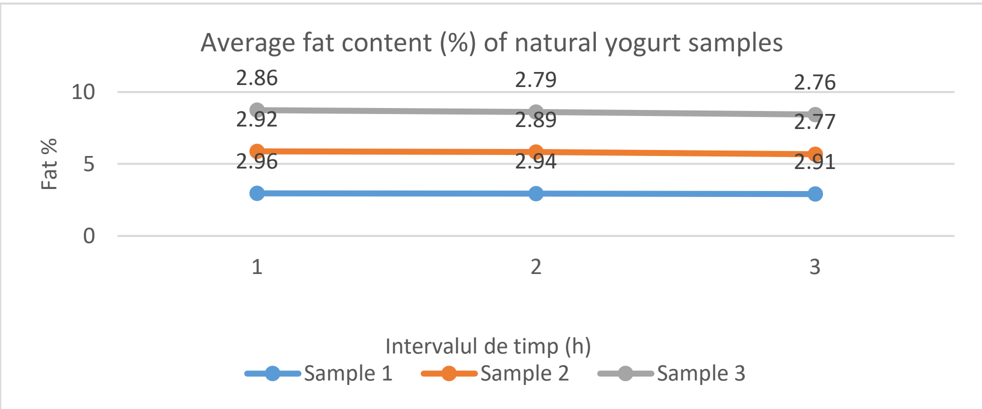

Figure 3. Average fat content (%) of natural yogurt samples

Average fat content of analyzed samples at the end of fermentation ranged between 2.76 % and 2.91 %, indicating an appropriate level for this type of yogurt classified as full-fat. Fat content analysis standards for yogurt follow SR EN ISO 1211:2010. These results indicate that the fermentation process used allowed the fat content to be maintained within the recommended limits, thus contributing to the production of a product with appropriate nutritional characteristics and acceptable quality for consumers.

CONCLUSIONS

The study evaluated the effect of different concentrations of mixed starter cultures of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* on the physicochemical properties of natural yogurt. Titratable acidity increased progressively during fermentation, while pH decreased accordingly, indicating the activity of lactic bacteria. Different proportions of starter cultures influenced the texture and syneresis of the product, with a higher proportion of *Streptococcus thermophilus* contributing to a creamier and more stable consistency.

The results allow the identification of the optimal starter culture concentration to obtain yogurt with a balance between acidity, texture, and stability. Depending on consumption preferences, natural yogurt can be served in various combinations: with nuts, hazelnuts, almonds, or different fruit jams, providing a healthy and complex product.

These findings provide useful insights for both industrial production and homemade yogurt preparation, highlighting the importance of starter culture management in achieving desirable sensory and physicochemical characteristics. Moreover, understanding the role of specific bacterial strains can guide further research on enhancing the nutritional and functional properties of fermented dairy products.

ACKNOWLEDGMENT

The author wishes to express gratitude to all who contributed to the completion of this study, especially for their guidance, technical support, and encouragement throughout the research process.

REFERENCES

Amani, E., Eskandari, M.H., & Shekarforoush, S. 2017. The effect of proteolytic activity of starter cultures on technologically important properties of yogurt. *Food Sci. Nutr.*, 5, 525–537. doi: 10.1002/fsn3.427.

Arioli, S., Scala, G.D., Remagni, M.C., Stuknyte, M., Colombo, S., & Guglielmetti, S. 2017. Streptococcus thermophilus urease activity boosts L. delbrueckii subsp. bulgaricus homolactic fermentation. *Int. J. Food Microbiol.*, 247, 55–64. doi: 10.1016/j.ijfoodmicro.2016.01.006.

Bibi, A., Xiong, Y., Rajoka, M.S.R., Mehwish, H.M., Radicetti, E., Umair, M., Shoukat, M., Khan, M.K.I., & Aadil, R.M. 2021. Recent Advances in the Production of Exopolysaccharide (EPS) from Lactobacillus spp. and its application in the food industry: A Review. *Sustainability*, 13, 12429. doi: 10.3390/su132212429.

Carroll, A.L., Desai, S.H., & Atsumi, S. 2016. Microbial production of scent and flavor compounds. *Curr. Opin. Biotechnol.*, 37, 8–15. doi: 10.1016/j.copbio.2015.09.003.

Cheng, H. 2010. Volatile flavor compounds in yogurt: A review. *Crit. Rev. Food Sci. Nutr.*, 50, 938–950. doi: 10.1080/10408390903044081.

Dan, T., Chen, H., Li, T., Tian, J., Ren, W., & Zhang, H.P. 2019. Influence of Lactobacillus plantarum P-8 on fermented milk flavor and storage stability. *Front. Microbiol.*, 9, 3133. doi: 10.3389/fmicb.2018.03133.

FAO. 2003. Food Energy—Methods of Analysis and Conversion Factors. Food and Agriculture Organization of the United Nations; Rome, Italy. FAO Food and Nutrition Paper 77.

FAO. 2018. WHO Standard for Fermented Milks. Food and Agriculture Organization of the United Nations; Rome, Italy.

Hadjimbei, E., Botsaris, G., & Chrysostomou, S. 2022. Beneficial Effects of Yoghurts and Probiotic Fermented Milks and Their Functional Food Potential. *Foods*, 11, 2691. doi: 10.3390/foods11172691.

Khiaosa-Ard, R., Kaltenegger, A., Humer, E., & Zebeli, Q. 2022. Effect of inclusion of bakery by-products in the dairy cow's diet on milk fatty acid composition. *J. Dairy Res.*, 89, 236–242. doi: 10.1017/S0022029922000619.

Sigala-Robles, R., Santiago-López, L., Hernández-Mendoza, A., Vallejo-Córdoba, B., Mata-Haro, V., Wall-Medrano, A., & González-Córdova, A.F. 2022. Peptides, Exopolysaccharides, and Short-Chain Fatty Acids from Fermented Milk and Perspectives on Inflammatory Bowel Diseases. *Digestive Diseases and Sciences*, 67, 4654–4665. doi: 10.1007/s10620-022-07382-2.

***SR EN 6345:1995, Lapte şi produse lactate. Analiză senzorială.

***SR EN ISO 1211:2010, Lapte. Determinarea conţinutului de grăsime. Metoda gravimetrică (Metodă de referinţă).