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**IMPROVEMENT OF THE BIOLOGICAL VALUE OF FERMENTED  
DAIRY PRODUCTS WITH BIOACTIVE INGREDIENTS FROM FRUITS**

**253.02. - Technology of products of animal origin**

**Summary of the doctoral thesis in engineering sciences**

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The doctoral thesis and the summary can be reviewed in the library of the Technical University of Moldova and on the ANACEC website ([www.anacec.md](http://www.anacec.md)).

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## CONCEPTUAL POINTS OF THE RESEARCH

**Motivation for choosing this subject.** The food industry is currently facing major challenges related to the need to ensure a balanced, diverse, and safe diet, adapted to an increasingly informed and health-conscious consumer. The demand for functional, natural products with beneficial properties for the body is growing, and scientific research focuses on optimizing processing and preservation technologies to maintain the bioactive components and sensory qualities of the final products. Berry fruits, such as aronia, raspberry, and strawberry, represent a valuable source of antioxidants, particularly polyphenols, anthocyanins, flavonoids, and vitamin C, which contribute to protecting the body against oxidative stress and reducing the risk of chronic diseases (Tamang et al., 2020; Gouda et al., 2021). On the other hand, these fruits are highly perishable, and conventional preservation methods, especially thermal ones, often compromise their nutritional and functional quality. At the same time, scientific interest is directed toward identifying technological solutions that allow optimal incorporation of berry fruits into the dairy matrix while preserving their functional properties and the stability of sensitive compounds. An interdisciplinary approach, combining aspects of biochemistry, microbiology, and food engineering, is essential for developing value-added products and for establishing modern technological processes.

**Relevance and importance of the topic addressed.** Fermented dairy products, particularly yogurt, represent one of the most popular food categories worldwide. Due to their balanced content of proteins, lipids, minerals, and live lactic cultures, yogurt provides benefits for digestion and intestinal microbiota health (Taleb et al., 2016; Verruck et al., 2019; Wang et al., 2023). One direction of innovation also lies in the use of a goat - cow milk blend. Goat milk is more easily digestible, has a unique lipid profile, and exhibits potential hypoallergenic properties, while cow milk provides a firm texture and stable protein structure. The combination of the two types of milk allows the production of fermented dairy products with superior rheological and sensory properties (Caleja et al., 2016; Gomez-Gallego et al., 2018). In this regard, the present study addresses both a practical need of the food industry and a current scientific objective by providing a basis for the production of safe, nutritious, and functional products. Additionally, the research contributes to strengthening the knowledge base regarding the interactions between the bioactive compounds of fruits and the dairy matrix, an essential aspect for optimizing technological processes.

In the context of the priority directions for the development of science in the Republic of Moldova, the topic of this thesis fully aligns with the research lines of national strategic interest, focused on "Sustainable Agriculture, Food Security, and Food Safety". The scientific approach is consistent with the objectives of modernizing food technologies, aiming at the creation of

innovative products with superior biological and functional value. A significant portion of the research was conducted within the State Program through project 20.80009.5107.09: "Improving Food Quality and Safety through Biotechnology and Food Engineering" (2020 - 2023), under the scientific supervision of prof. dr. hab. Rodica Sturza, which ensured the integration of the obtained results into the national scientific circuit and their practical applicability for the food industry. In this way, the thesis contributes both to the theoretical foundation of modern solutions for the valorization of agri - food resources and to supporting sustainable development and the competitiveness of the Republic of Moldova.

**The purpose of the research:** is to obtain berry fruit purees preserved using a combined ultrasonication and sterilization method while maintaining a high antioxidant potential, and to use these purees in the production of yogurt with enhanced biological value.

**The general objective** of the work is the development and scientific establishment of innovative fruit preservation technologies for their use as functional ingredients in yogurts, aiming to improve the nutritional value, sensory characteristics, and stability of the final product.

To achieve the aim, the following **operational objectives** were formulated:

1. To investigate the influence of ultrasonication and sterilization conditions on the biological value and antioxidant potential of berry fruit purees, and to establish mathematical models for determining the optimal preservation conditions.
2. To determine the optimal ultrasonication and sterilization parameters affecting the sensory characteristics, physicochemical indicators (titratable acidity, pH, dry matter, ash, protein, fiber, sugar), biological value (*L*- ascorbic acid, total anthocyanin content), and antioxidant activity of berry fruit purees.
3. To determine the optimal ratio of the goat - cow milk mixture and to develop the technology for producing yogurt from the goat - cow milk mixture.
4. To investigate the influence of berry fruit puree in yogurt and the storage duration on sensory characteristics, physicochemical indicators, microbiological stability, biological value, antioxidant activity, and to establish a mathematical model for the quality of the final product.

**The research hypothesis** is that the combined ultrasonication and sterilization method allows the preservation of the high antioxidant potential of berry fruit purees and their use in the production of yogurt from a goat - cow milk mixture with enhanced biological value.

**Synthesis of the research methodology and justification of the chosen research methods.** The study was based on preservation methods using ultrasonication and sterilization, as well as their combination at different temperatures and durations. The HPLC method was applied for the characterization of berry fruit purees and yogurts made from a goat - cow milk mixture

with berry fruit purees. Antioxidant activity (DPPH and ABTS) was determined for both the berry fruit purees and the prepared yogurts. Sensory, microbiological, physicochemical, and textural analyses were also performed. Statistical processing methods and mathematical modeling of the experimental results were applied.

**The theoretical importance and scientific innovation of the work** consist in determining the optimal preservation conditions for berry fruit purees using a combined thermal treatment of ultrasonication and sterilization, which allows the preservation of biological value and antioxidant activity; identifying the optimal preservation conditions for standardized high - antioxidant berry fruit purees through combined ultrasonication and sterilization; providing the rationale for using a goat - cow milk mixture in yogurt production

**Theoretical significance:** for the first time, the kinetics of the preservation method using combined thermal ultrasonication and sterilization of berry fruit purees at different temperatures and durations was modeled; the optimal preservation conditions of the thermal ultrasonication and sterilization treatment were established with respect to physicochemical indicators, biological value, and antioxidant activity of the berry fruit purees; technologies for producing yogurt from a goat - cow milk mixture were developed, in which berry fruit purees were used as a ingredient to enhance the biological value of the final product.

**Approval of the work at national and international scientific forums.** The main results of the thesis were presented at a series of national and international scientific conferences and symposium.

**Publications on the topic of the thesis.** The research results and issues addressed in the thesis have been published in 19 scientific works, including a chapter in a collective monograph, 7 scientific articles, a short - term invention patent, and 12 articles in proceedings and abstracts presented at national and international scientific events.

**Summary of the thesis chapters.** The thesis is presented on 132 typed pages and includes the following sections: abstracts in romanian, english, and russian; introduction; 4 chapters; conclusions and recommendations; and a bibliography with 256 sources. The work is illustrated with 35 tables and 49 figures, and includes 2 appendices.

**Keywords:** goat milk, cow milk, fruits, starter culture, yogurt, fermentation, acidity, lactic bacteria, protein, viscosity, texture, firmness, syneresis, antioxidants, stability.

## **THESIS CONTENT**

### **1. INNOVATIVE FRUIT PRESERVATION TECHNOLOGIES FOR ENHANCING YOGURT QUALITY**

The first chapter of the thesis presents a literature review on the application of modern preservation technologies for berry fruits and their integration into fermented dairy products with high biological value. Incorporating berry fruit purees into fermented yogurts ensures harmonious integration, resulting in products with high physicochemical stability, persistent natural color, and superior functional value. Thus, the combination of innovative fruit preservation technologies, the use of mixed milk, and the addition of berry purees represents a promising direction for the development of functional yogurts with nutritional benefits and a positive impact on consumer health.

### **2. RESEARCH MATERIALS AND METHODS**

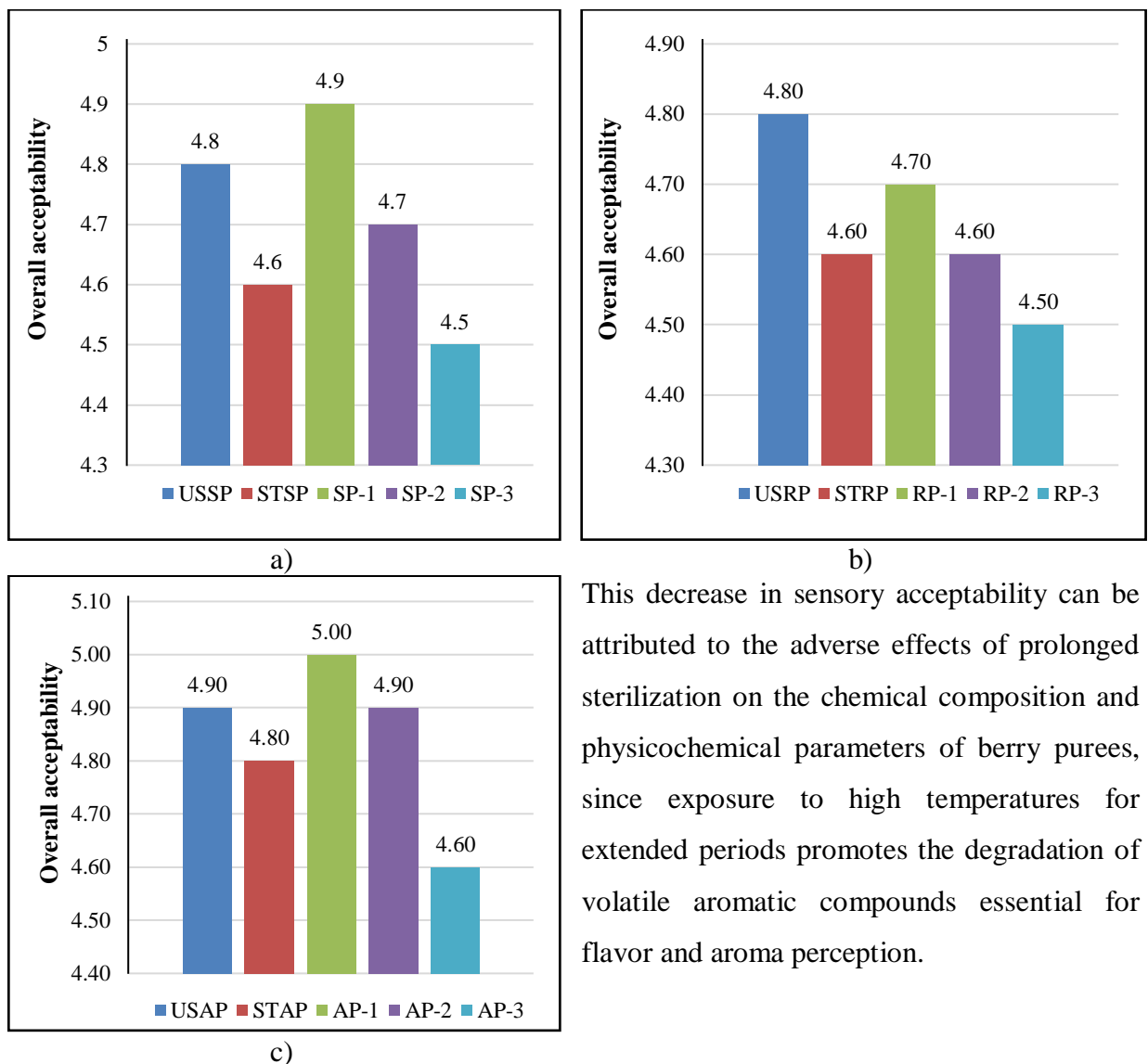
Chapter 2 provides a detailed description of the raw materials used in the research, the methods applied for the preparation and preservation of berry fruit purees, as well as the methodology for analyzing the fermented yogurts obtained through the incorporation of these plant ingredients. The experiments utilized berry fruits (aronia, strawberry, raspberry), cow and goat milk, and specific reagents for physicochemical, sensory, and microbiological analyses. The preservation of the purees was carried out using a combined method: ultrasonication at  $70 \pm 1^\circ\text{C}$  for  $12 \pm 2$  minutes, followed by sterilization at  $105^\circ\text{C}$  for 90 - 290 seconds. The parameters were varied to evaluate the impact on bioactive compounds (*L*- ascorbic acid, anthocyanins), antioxidant activity, sensory characteristics, and microbiological stability. Experimental data were expressed as means  $\pm$  standard error of the mean (SEM), calculated from three independent determinations. Statistical analysis was performed using Microsoft Excel 2007 (Microsoft Corporation, Redmond, WA, USA), applying one - way ANOVA to identify significant differences between groups, followed by Tukey's post - hoc test. Differences were considered significant at  $p \leq 0.05$ .

### **3. OPTIMIZATION OF BERRY PROCESSING THROUGH INNOVATIVE TECHNIQUES**

This chapter examined the application of thermal treatments using ultrasound and the combination of thermal treatment through ultrasonication and sterilization to extend the shelf life of fruit purees, ensuring high microbiological stability, improving sensory characteristics, physicochemical indicators, and the content of biologically active compounds.

### 3.1 Analysis of quality indicators of berry fruit purees

**3.1.1 Sensory characteristics of berry fruit purees.** The sensory evaluation of berry fruit purees highlighted the superiority of the combined thermal treatment using ultrasonication and sterilization compared to the treatments applied individually (either sterilization or ultrasonication alone), as shown in Figure 1 (a, b, c). For purees preserved by ultrasonication alone, the mean scores of overall acceptability were 4.8 for strawberry (USSP) and raspberry (USRP) purees, and 4.9 for aronia puree (USAP). In contrast, purees subjected to the combined thermal treatment, which included ultrasonication followed by sterilization for 135 seconds, achieved higher sensory scores: 4.9 for sample SP-1 (strawberry), 4.7 for RP-1 (raspberry), and 5.0 for AP-1 (aronia). Berry fruit purees scored 4.5 for samples SP-3 and RP-3, and 4.6 for AP-3.



**Fig. 1 Overall acceptability of berry purees**  
a) strawberry purees, b) raspberry purees,  
c) aronia purees

### 3.1.2 Antioxidant potential and microbiological indicators of berry fruit purees.

This decrease in sensory acceptability can be attributed to the adverse effects of prolonged sterilization on the chemical composition and physicochemical parameters of berry purees, since exposure to high temperatures for extended periods promotes the degradation of volatile aromatic compounds essential for flavor and aroma perception.



Table 1 highlights the antioxidant potential of the berry purees by determining the *L*-ascorbic acid content under various heat treatment conditions. The results indicate that the combined thermal treatment - consisting of ultrasonication at  $70 \pm 1^\circ\text{C}$  for 12 - 16 min, followed by sterilization at  $105^\circ\text{C}$  for 135 s - ensures better preservation of *L*-ascorbic acid in all samples compared to the regime with extended sterilization of 290 s. The highest concentrations of *L*-ascorbic acid were recorded in samples SP-2 (39.53 mg/100 g) and RP-2 (18.46 mg/100 g), while sample AP-2 exhibited a significantly lower content (7.70 mg/100 g). Comparative analysis of *L*-ascorbic acid losses revealed decreases of 21.07% in strawberry purees, 31.77% in raspberry purees, and 68.23% in aronia purees as a result of prolonged sterilization. The degradation of *L*-ascorbic acid during thermal processing can be explained by thermodegradative mechanisms involving oxidation and thermal decomposition. In particular, prolonged exposure to high temperatures promotes the breakdown of the chemical bonds of *L*-ascorbic acid, leading to the formation of compounds that are inactive from an antioxidant standpoint. The substantial differences in the degree of degradation among the purée types can be attributed to the variability of the food matrix and the specific chemical composition of each fruit, which influence the stability of *L*-ascorbic acid during thermal processing. Thus, the aronia matrix appears to be more susceptible to degradation, as evidenced by the more pronounced reduction in *L*-ascorbic acid concentration observed in sample AP-2.

The highest total anthocyanin content was observed in sample AP-2, with 150.51 mg/g, followed by samples RP-2 (71.98 mg/100 g) and SP-2 (37.72 mg/100 g). However, prolonging the sterilization time led to a significant decrease in extractable anthocyanin content, with the lowest values recorded in samples AP-3 (104.59 mg/100 g), RP-3 (61.62 mg/100 g), and SP-3 (28.21 mg/100 g). The reduction in anthocyanin content can be attributed both to thermodegradative mechanisms and to specific technological effects associated with the sterilization process. Thermally, anthocyanins are susceptible to chemical transformations, including dehydration and conversion into unstable chalcone forms, resulting in color loss and diminished antioxidant capacity. From a technological perspective, prolonged sterilization times and elevated temperatures may induce structural modifications within the fruit matrix, such as pectin gelation and protein denaturation, which affect the accessibility and stability of anthocyanins. Thus, the decrease in anthocyanin content in berry purees subjected to sterilization is the outcome of a combination of direct thermal degradation of pigments and multiple technological effects that alter the structural and compositional integrity of the food matrix, ultimately influencing the sensory quality and nutritional value of the final product.

**Table 1. Effect of sterilization on the antioxidant potential and microbiological indicators of berry purees obtained by ultrasonication**

Sample	Ultrasonication		Sterilization		L- ascorbic acid content, mg/100 g	Total anthocyanin content, mg/100 g	Antioxidant activity		TNFAnM, log CFU/g	Yeasts, molds, log CFU/g
	t, °C	τ, min	t, °C	τ, s			DPPH, mg TE/100 g	ABTS, mg TE/100 g		
Strawberry purees										
USSP	70	12	-	-	35.37±0.56	32.03±0.60	6.25±0.13	14.56±0.24	3.60	1.84
STSP	-	-	105	290	33.17±0.52	30.76±0.62	6.17±0.14	14.12±0.25	3.48	1.78
SP-1	70	12	105	90	31.06±0.53	31.80±0.63	6.83±0.15	12.97±0.21	3.43	1.30
SP-2				135	39.53±0.51	37.72±0.60	6.97±0.10	15.21±0.36	3.00	<1
SP-3				290	30.37±0.51	28.21±0.63	5.35±0.05	10.61±0.26	2.48	<1
Raspberry purees										
USRP	70	12	-	-	15.16±0.52	68.34±0.62	5.68±0.02	10.02±0.14	3.48	1.90
STRP	-	-	105	290	13.24±0.51	66.39±0.63	5.42±0.06	8.69±0.15	3.45	1.84
RP-1	70	12	105	90	12.98±0.52	67.11±0.66	5.75±0.05	7.52±0.18	3.00	1.30
RP-2				135	18.46±0.54	71.98±0.65	6.32±0.15	12.32±0.27	2.48	<1
RP-3				290	10.45±0.53	61.62±0.61	5.31±0.11	4.65±0.04	2.00	<1
Aronia purees										
USAP	70	12	-	-	5.30±0.52	132.78±0.62	16.89±0.34	32.87±0.67	3.48	1.69
STAP	-	-	105	290	4.80±0.54	125.18±0.63	16.19±0.34	32.19±0.51	3.00	1.60
AP-1	70	12	105	90	4.38±0.58	129.39±0.64	15.25±0.31	31.83±0.65	2.90	<1
AP-2				135	7.70±0.55	150.51±0.64	17.54±0.35	39.72±0.95	2.30	<1
AP-3				290	3.14±0.53	104.59±0.63	10.89±0.22	27.68±0.79	2.00	<1

\* Note: USSP - ultrasonicated strawberry puree, STSP - sterilized strawberry puree, SP-1, SP-2, and SP-3 - strawberry puree sterilized for 90, 135, and 290 s, respectively; USRP - ultrasonicated raspberry puree, STRP - sterilized raspberry puree, RP-1, RP-2, and RP-3 - raspberry puree sterilized for 90, 135, and 290 s, respectively; USAP - ultrasonicated aronia puree, STAP - sterilized aronia puree, AP-1, AP-2, and AP-3 - aronia puree sterilized for 90, 135, and 290 s, respectively; TTU - thermal treatment by ultrasonication; TTS - thermal treatment by sterilization. Values represent the mean of three independent determinations ± standard deviation,  $P \leq 0.05$

The samples that exhibited the highest values for *L*- ascorbic acid and anthocyanins (SP-2, RP-2, AP-2) corresponded to the maximum AA<sub>DPPH</sub> and AA<sub>ABTS</sub> values, whereas prolonged sterilization (SP-3, RP-3, AP-3) was associated with a significant decrease in these indicators, both quantitatively and functionally. These trends suggest that structural modifications of anthocyanins and oxidative degradation of *L*- ascorbic acid are synergistic processes contributing to the loss of antioxidant activity. The microbiological indicators show that the total number of aerobic mesophilic microorganisms (TNFAnM) in all analyzed berry purée samples remained below the maximum permissible limit for this type of food product, i.e., 5 log CFU/g, according to current regulations. It was found that combined thermal treatments integrating ultrasonication ( $70 \pm 1^\circ\text{C}$  for 12 min) and sterilization at  $105^\circ\text{C}$  were the most effective in reducing microbial load. The lowest TNFAnM values were recorded for the combined thermal treatment with extended sterilization time of 290 s. Specifically, sample SP-3 showed a contamination level of 2.48 log CFU/g, while samples RP-3 and AP-3 exhibited a minimum value of 2.00 log CFU/g. Regarding the presence of yeasts and molds, the results did not indicate any exceedance of the permissible microbiological limits, confirming the effectiveness of the applied treatments.

### **3.1.3 Physicochemical indicators of berry fruit purees.**

Table 2 summarizes the effects of thermal treatments on the physicochemical indicators of berry purees. The results showed that titratable acidity and pH values did not experience significant changes ( $p < 0.05$ ). Titratable acidity ranged from 0.79 to 0.85%, while pH values varied between 4.01 and 4.27, depending on the type of fruit puree. The highest dry matter content was 21.6 °Brix in aronia purée (AP-2), whereas the other samples showed values between 8.4 and 8.9 °Brix. Regarding ash content, aronia purees exhibited the highest values (0.77%). The highest protein contents were recorded in aronia purees (4.11%), raspberry purees (1.25%), and strawberry purees (0.70%). Total fiber content was highest in raspberry puree (7.78%), while strawberry and aronia purees had lower values, reflecting the specific tissue structure of the fruits and their composition in cellulose, hemicellulose, and pectin.

Additionally, total sugars were highest in raspberry purees (11.90%), followed by strawberry and aronia. Statistical analysis confirmed that individual ultrasonication treatments or combined treatments with sterilization did not cause significant changes in the physico - chemical parameters of berry purees ( $p > 0.05$ ). These results suggest that the applied thermal treatments have minimal impact on the physical characteristics of berry purees, thereby highlighting their technological and nutritional potential for applications in the food industry.

**Table 2. Effect of ultrasonication and sterilization on the physicochemical parameters of berry purees**

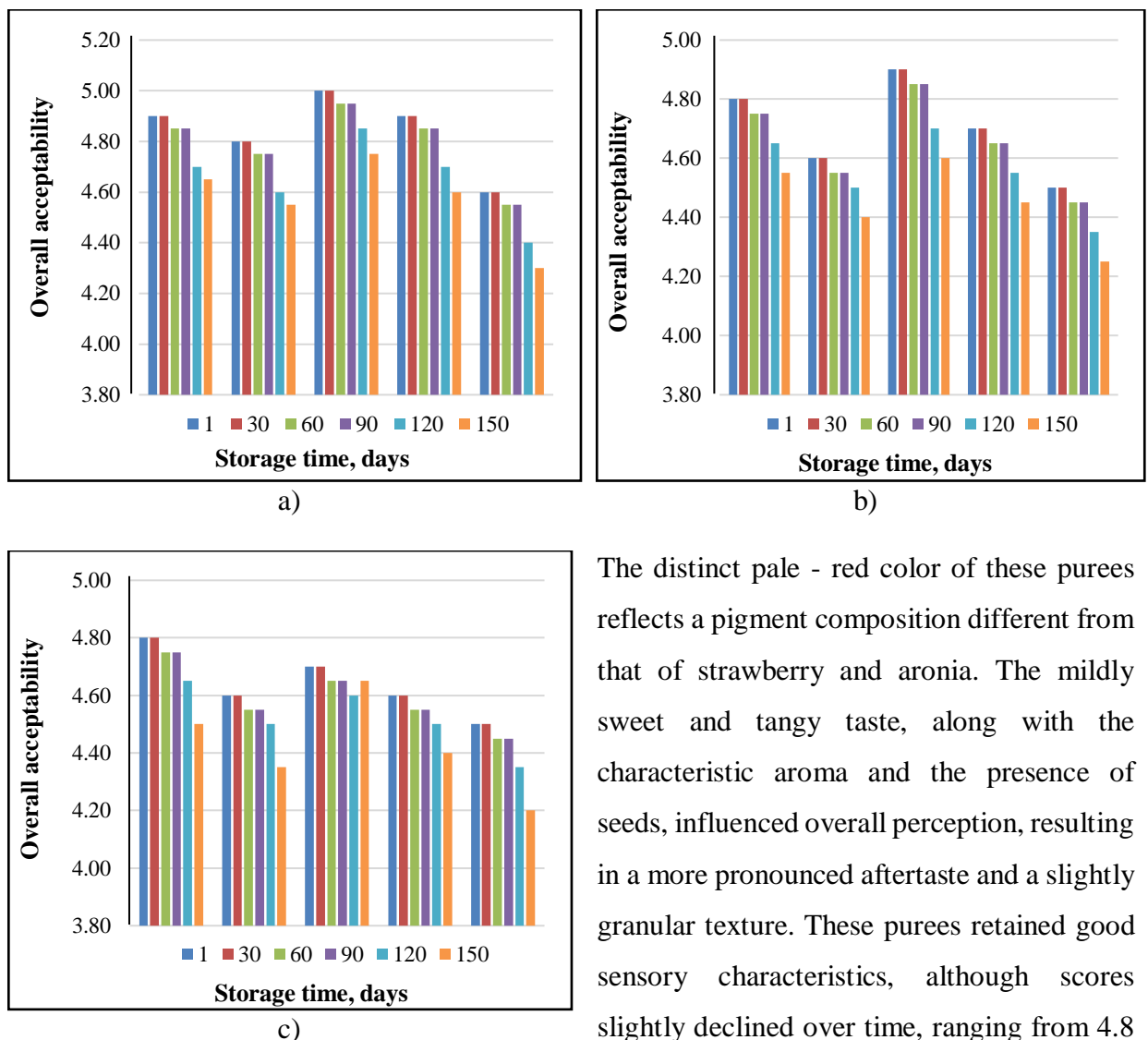
Sample	Ultrasonication		Sterilization		Titratable acidity, %	pH	Dry matter content, °Brix	Ash content, %	Protein content, %	Total fiber content, %	Total sugar content, g/100g
	t, °C	τ, min	t, °C	τ, s							
Strawberry purees											
USSP	70	12	-	-	0.81±0.06 * m.a.	4.24±0.01	8.3±0.1	0.64±0.01	0.67±0.02	1.98±0.04	9.71±0.05
STSP	-		105	290	0.80±0.06 * m.a.	4.23±0.02	8.1±0.2	0.63±0.01	0.65±0.02	1.96±0.02	9.67±0.04
SP-1	70	12	105	90	0.82±0.03 * m.a.	4.26±0.02	8.0±0.2	0.65±0.02	0.68±0.01	1.95±0.02	9.69±0.04
SP-2				135	0.84±0.04 * m.a.	4.27±0.01	8.4±0.1	0.68±0.01	0.70±0.02	2.03±0.04	9.74±0.03
SP-3				290	0.82±0.05 * m.a.	4.25±0.01	8.2±0.1	0.66±0.02	0.64±0.01	1.93±0.04	9.65±0.04
Raspberry purees											
USRP	70	12	-	-	0.80±0.05 ** a.a.	4.19±0.02	8.7±0.2	0.62±0.02	1.21±0.01	7.75±0.02	11.81±0.03
STRP	-		105	290	0.79±0.06 ** a.a.	4.17±0.02	8.6±0.2	0.60±0.01	1.18±0.01	7.71±0.02	11.77±0.04
RP-1	70	12	105	90	0.82±0.02 ** a.a.	4.16±0.01	8.8±0.1	0.63±0.01	1.22±0.02	7.73±0.04	11.76±0.04
RP-2				135	0.83±0.04 ** a.a.	4.21±0.02	8.9±0.1	0.66±0.02	1.25±0.02	7.78±0.04	11.90±0.04
RP-3				290	0.81±0.07 ** a.a.	4.20±0.01	8.5±0.2	0.64±0.02	1.23±0.01	7.69±0.02	11.74±0.05
Aronia purees											
USAP	70	12	-	-	0.82±0.05 * m.a.	4.04±0.02	21.4±0.2	0.73±0.01	4.08±0.02	1.46±0.04	6.94±0.04
STAP	-		105	290	0.80±0.04 * m.a.	4.02±0.01	21.2±0.1	0.72±0.02	4.06±0.01	1.42±0.02	6.90±0.03
AP-1	70	12	105	90	0.82±0.05 * m.a.	4.01±0.01	21.1±0.2	0.74±0.01	4.05±0.01	1.40±0.02	6.93±0.04
AP-2				135	0.85±0.07 * m.a.	4.06±0.02	21.6±0.1	0.77±0.02	4.11±0.01	1.50±0.04	6.98±0.05
AP-3				290	0.83±0.03 * m.a.	4.03±0.01	21.0±0.1	0.75±0.01	4.04±0.02	1.39±0.02	6.75±0.04

\*Note: USSP - ultrasonicated strawberry puree, STSP - sterilized strawberry puree, SP-1, SP-2, and SP-3 - strawberry puree sterilized for 90, 135, and 290 s, respectively; USRP - ultrasonicated raspberry puree, STRP - sterilized raspberry puree, RP-1, RP-2, and RP-3 - raspberry puree sterilized for 90, 135, and 290 s, respectively; USAP - ultrasonicated aronia puree, STAP - sterilized aronia puree, AP-1, AP-2, and AP-3 - aronia puree sterilized for 90, 135, and 290 s, respectively; TTU - thermal treatment by ultrasonication; TTS - thermal treatment by sterilization; \* m.a. - malic acid; \*\* a.a. - ascorbic acid.

Values represent the mean of three independent determinations ± standard deviation,  $P \leq 0.05$

### 3.2 Effect of storage duration on quality indicators of berry fruit purees

**3.2.1 Sensory characteristics of berry fruit purees during storage.** The sensory evaluation carried out during storage, illustrated in Figure 2 (a, b, c), highlights the differences between strawberry, raspberry, and aronia purees. Strawberry purees consistently received the highest acceptability scores throughout the storage period. The sweet, pleasant taste with lower acidity compared to the other samples was a key factor in maintaining consumer preference. The creamy and less fibrous texture, along with a sweet aroma and uniform appearance, contributed to the superior sensory stability of this type of puree. Raspberry purees maintained good sensory characteristics, although with a slight decline in scores over time, ranging from 4.8 to 4.3 points.



**Fig. 2 Overall acceptability of berry purees during storage**  
**a) strawberry purees, b) raspberry purees, c) aronia purees**

The distinct pale - red color of these purees reflects a pigment composition different from that of strawberry and aronia. The mildly sweet and tangy taste, along with the characteristic aroma and the presence of seeds, influenced overall perception, resulting in a more pronounced aftertaste and a slightly granular texture. These purees retained good sensory characteristics, although scores slightly declined over time, ranging from 4.8 to 4.3 points. Aronia purees were the least appreciated in terms of taste, with scores ranging from 4.7 to 4.2 points, exhibiting

pronounced astringency due to their high tannin content. The less sweet flavor and dry aftertaste are typical characteristics of aronia, which may affect the product's overall acceptability.

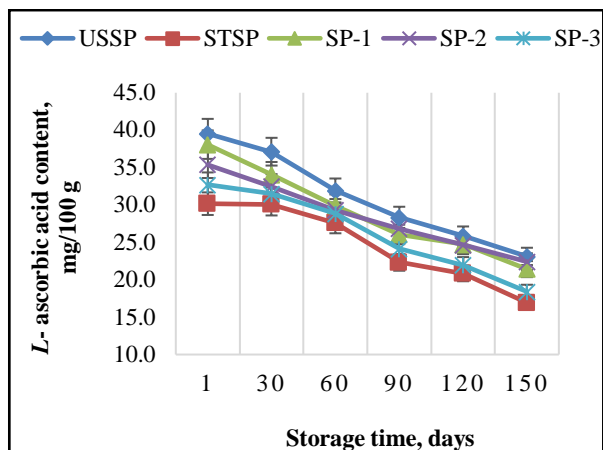
Additionally, the intense violet color of aronia purée indicates a high concentration of anthocyanins, bioactive compounds that, while beneficial for health, can contribute to astringency. These factors explain the lower sensory scores compared to strawberry and raspberry purees. Thus, the differences in the evolution of sensory characteristics during storage are directly influenced by the specific chemical composition of each fruit, as well as by how the structure and bioactive compounds respond to thermal treatments and storage conditions.

### **3.2.2 Biological value of berry fruit purees during storage.**

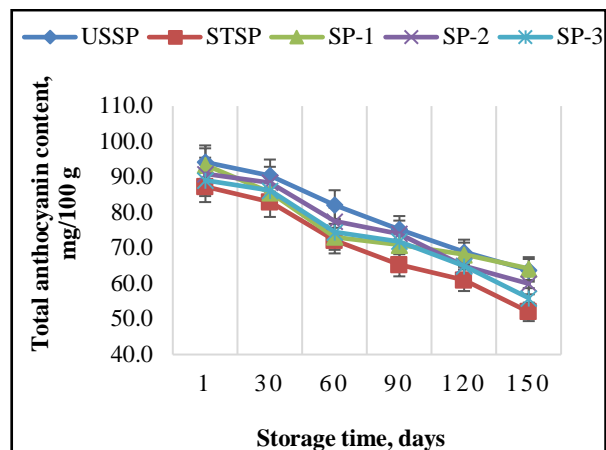
The *L*- ascorbic acid content, shown in Figure 3 (a, b, c), indicates a significant decrease, with variations depending on the type of purée and the applied treatment method. Aronia purees exhibited the most pronounced decline in *L*- ascorbic acid. For example, thermal sterilization led to a reduction of 69.50%, from 5.87 mg/100 g on day 1 to 1.39 mg/100 g after 150 days (sample STAP). Ultrasonication resulted in a 55.94% decrease, while combined treatments showed reductions ranging from 62.60% to 64.41%, depending on the treatment duration. Raspberry purees showed a similar trend, with substantial decreases in *L*- ascorbic acid. Thermal sterilization caused a 63.04% reduction, while ultrasonication decreased the content by 55.94%. Combined treatments resulted in a decline between 55.66% and 57.14%, correlated with the process duration. Strawberry purees demonstrated a comparatively lower rate of *L*- ascorbic acid loss. Sterilization reduced the content by 43.88%, and ultrasonication by 41.51%.

The analysis of total anthocyanin content, presented in Figure 4 (a, b, c), shows that strawberry purees experienced the most significant losses, with a decrease of 40.42% under sterilization and 31.25% under ultrasonication. Combined thermal treatment resulted in reductions of up to 37.25%. In contrast, aronia purees retained most of their total anthocyanin content, with minimal losses (below 23%).

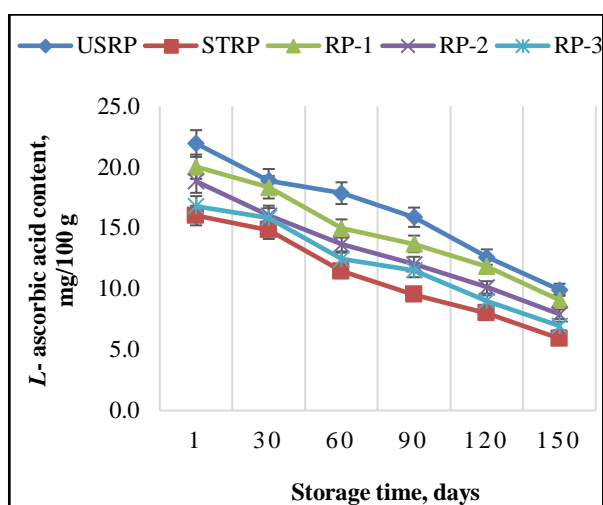
These trends are influenced by complex chemical and physical factors: the degradation of *L*- ascorbic acid is accelerated by oxidation, which generates hydrogen peroxide and other reactive species that break down antioxidants (Sulaiman et al., 2015; Aguilar et al., 2017). Moreover, the duration of exposure to thermal treatments has a more pronounced impact than temperature alone, promoting oxidative processes and polymerization of phenolic compounds, which lead to a decrease in antioxidant activity.



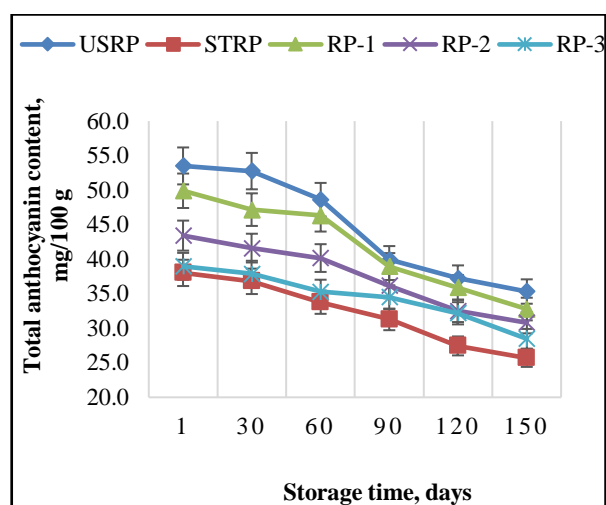
a)



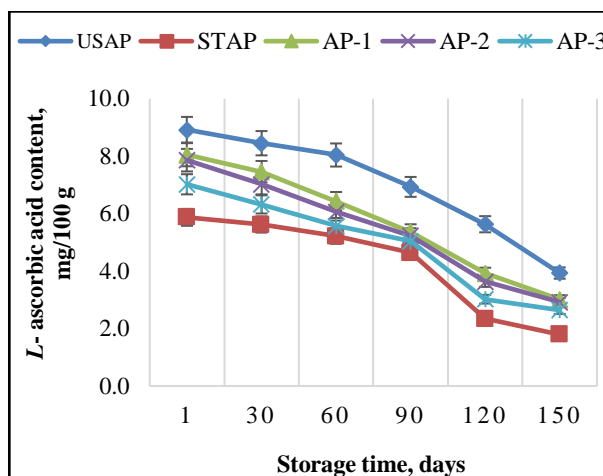
a)



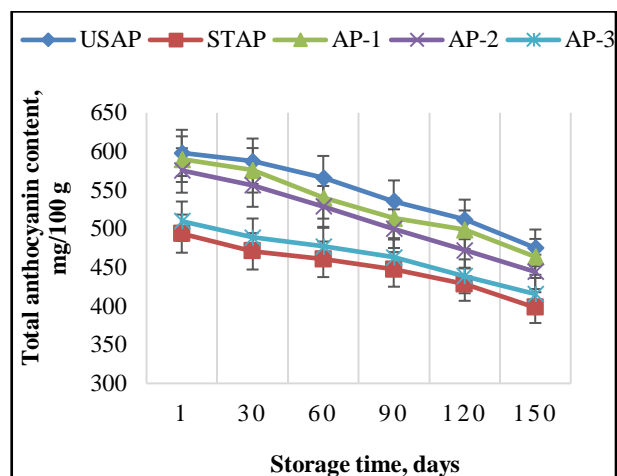
b)



b)



c)



c)

**Fig. 3 Evolution of *L*- ascorbic acid content in berry purees during storage**  
a) strawberry purees, b) raspberry purees, c) aronia purees

**Fig. 4 Evolution of total anthocyanin content in berry purees during storage**  
a) strawberry purees, b) raspberry purees, c) aronia purees

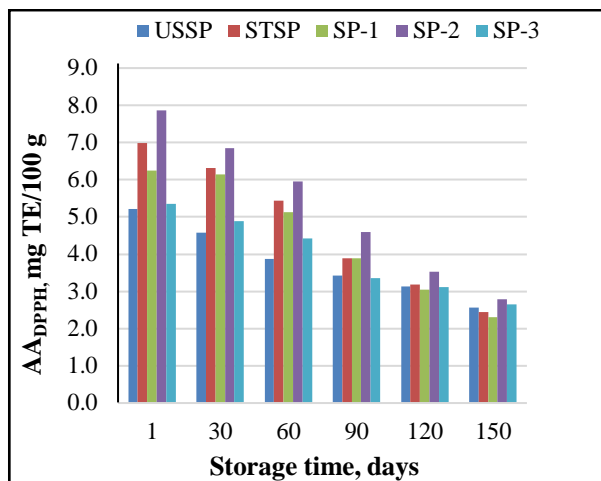
### 3.2.3 Antioxidant activity of berry fruit purees during storage.

The results presented in Figure 5 (a, b, c, a', b', c') indicate that purees subjected to sterilization exhibited higher antioxidant activity by DPPH (54.58%) compared to those obtained through ultrasonication (46.89%). Notably, aronia purees showed the lowest rate of  $AA_{DPPH}$  decline, which can be attributed to their initially high concentrations of anthocyanins and *L*-ascorbic acid, providing greater resistance to degradation. For example, in sample STAP, thermal sterilization led to only a 48.36% decrease in  $AA_{DPPH}$ , where as ultrasonication caused a 43.34% reduction.

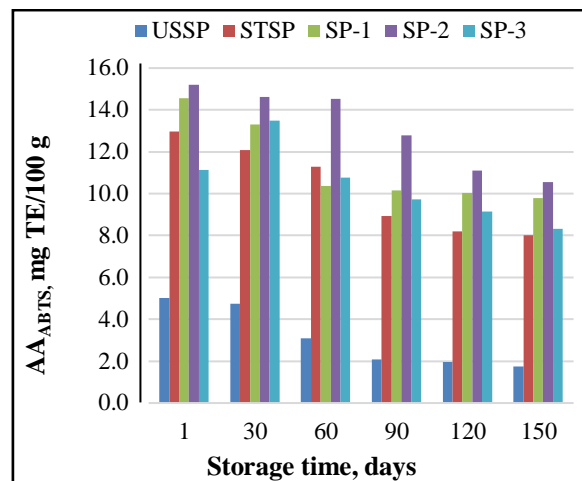
This higher stability can be ascribed to the relatively robust anthocyanin content in aronia and the synergistic interactions among bioactive compounds that slow down oxidative degradation. Raspberry purees showed a similar decreasing trend, although with a slightly higher rate of decline, particularly after thermal sterilization, indicating greater sensitivity of raspberry antioxidant compounds to processing and storage conditions. In comparison, strawberry purees exhibited the highest  $AA_{DPPH}$  reduction, reaching up to 64.89% after sterilization, reflecting lower stability of anthocyanins and *L*-ascorbic acid in these berry purees. These differences highlight the importance of the nature and concentration of bioactive compounds specific to each type of puree. Furthermore, the analysis of antioxidant activity using the ABTS method ( $AA_{ABTS}$ ) confirmed the trends observed with DPPH, but with higher values, which can be attributed to the differing sensitivities of the two methods to various antioxidants present in berry purees. It is well known that the ABTS method can quantify a broader spectrum of both water- and fat-soluble antioxidants compared to DPPH, which explains why the antioxidant activity measured by ABTS was approximately 12.45% higher. The progressive decline of  $AA_{ABTS}$  during storage is closely related to the degradation of *L*-ascorbic acid and phenolic compounds, a process accelerated by the duration and conditions of the combined thermal treatment.

Thus, it can be observed that berry purees exhibit different behaviors regarding antioxidant stability during storage. Higher antioxidant activity values in purée samples subjected to sterilization suggest partial protection of bioactive compounds, despite the losses recorded. At the same time, the differences among the samples highlight the role of the initial anthocyanin and *L*-ascorbic acid content in maintaining antioxidant activity. The evolution of these parameters throughout storage confirms the trend of a progressive decline in antioxidant capacity. Therefore, the results clearly illustrate how the applied treatments influence the stability of functional compounds in the studied purees.

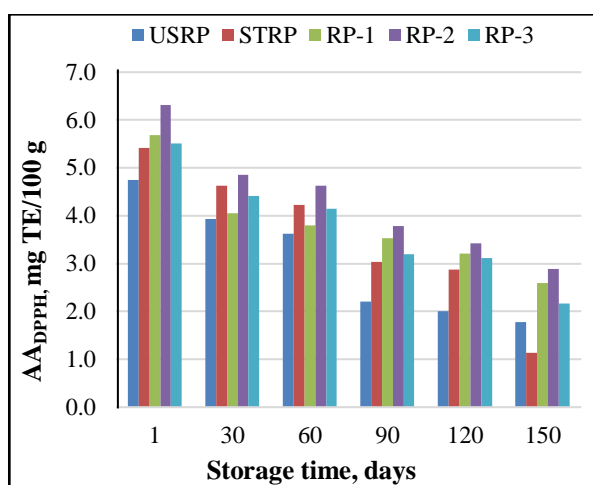




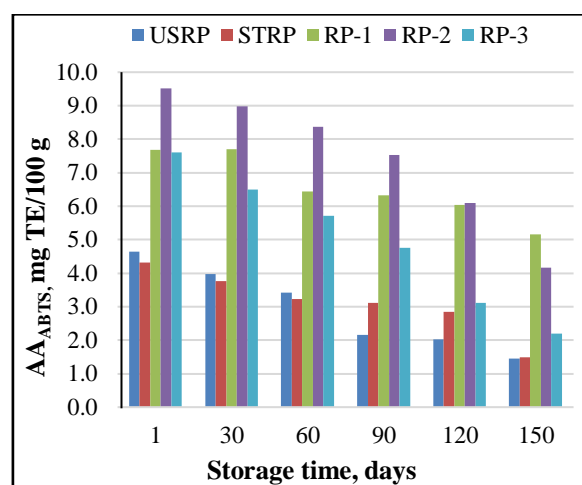
a)



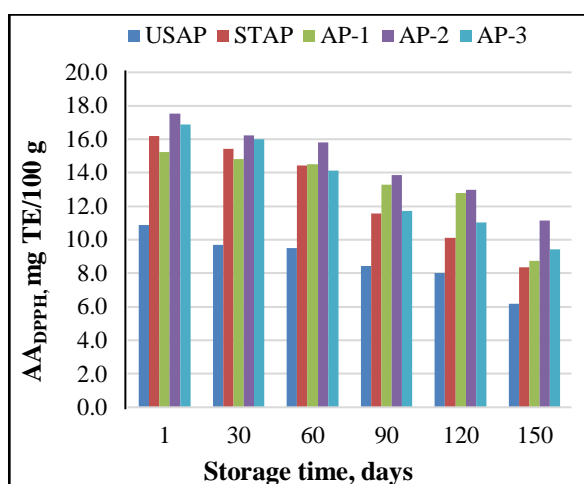
a')



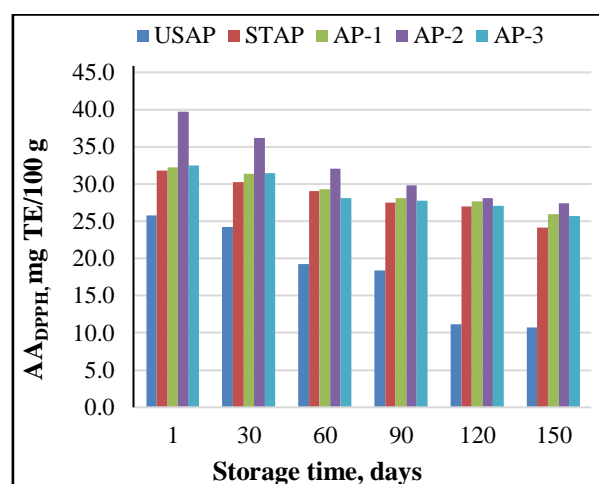
b)



b')



c)



c')

**Fig. 5 Evolution of DPPH (a, b, c) and ABTS (a', b', c') antioxidant activity in berry purees during storage**

**a) strawberry purees, b) raspberry purees, c) aronia purees**

### 3.2.4 Microbiological indicators of berry fruit purees during storage.

The evaluation of the impact of thermal treatments on the microbiological indicators of berry purees during storage (Table 3) showed a positive effect in reducing the total number of facultative anaerobic microorganisms (TNFAnM), yeasts, and molds. In contrast, purees preserved by ultrasonication (USSP, USRP, USAP) exhibited higher microbial counts, indicating a lower efficiency of this method in controlling spoilage microbiota. For example, combining ultrasonication with sterilization (sample SP-2) resulted in a bacterial reduction of approximately 3 log CFU/g, whereas ultrasonication alone achieved a higher inactivation, around 4 log CFU/g in certain samples (USRP and USAP). During storage under limited - oxygen conditions (twist - off jars), both ultrasonicated and combined - treatment samples showed a decrease in microorganisms, yeasts, and molds, maintaining levels below critical microbial contamination thresholds.

At the end of the 150- day storage period, combined thermal treatments maintained a low level of TNFAnM, with reductions of less than 2 log CFU/g compared to initial values, while yeasts and molds remained below the detection limit, indicating satisfactory microbiological stability. Berry purees preserved by ultrasonication did not show significant changes during storage, highlighting the limitations of this method in ensuring long - term microbiological safety. Furthermore, the residual microbiota remained within sanitary limits, with microorganism counts below 1 log CFU/g, considered acceptable for food safety and the stability of berry purees during storage. Tests confirmed the absence of microbial growth under anaerobic conditions, demonstrating that combined thermal treatments are effective for maintaining microbiological quality. In addition, pathogenic microbiota, including dangerous non - spore - forming bacteria such as *Escherichia coli*, *Salmonella*, and *Staphylococcus*, were not detected in the berry purees, owing to their sensitivity to the applied thermal treatments.

These results highlight the importance of combining thermal treatments with ultrasonication not only for reducing microbial populations but also for preserving the integrity of bioactive compounds in the purees. The achieved microbiological stability suggests that the applied methods prevent both organoleptic deterioration and the loss of functional properties over the long term.

Furthermore, statistical analysis of the data confirmed significant differences between samples subjected to combined treatment and those treated with ultrasonication, emphasizing the superior efficiency of the integrated strategy. The obtained results support the development of standardized preservation protocols applicable at an industrial scale and provide a basis for microbiological risk assessment and the implementation of food safety measures.

**Table 3. Microbiological indicators of berry purees during storage**

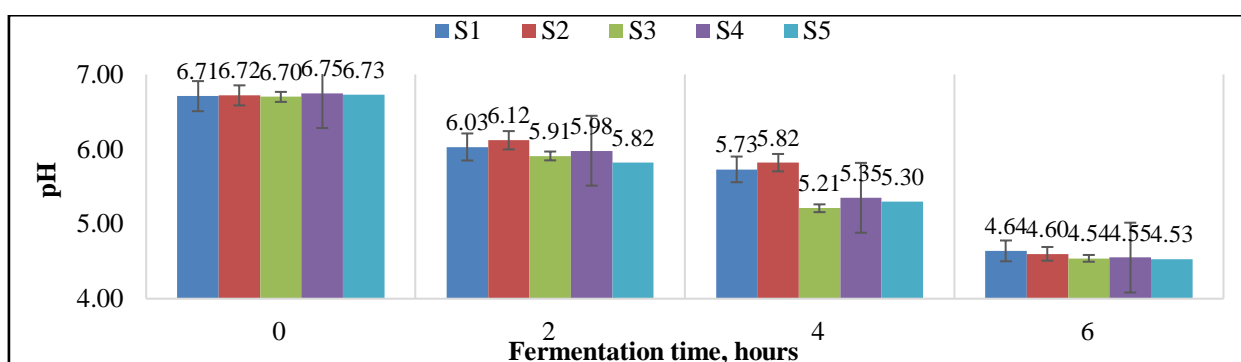
Sample	Ultrasonication		Sterilisation		TNFAnM, log CFU/g						Yeasts, molds, log CFU/g					
					Storage time, days											
	t, °C	τ, min	t, °C	τ, s	1	30	60	90	120	150	1	30	60	90	120	150
Strawberry purees																
USSP	70	12	-	-	3.60	3.95	3.95	4.00	4.00	4.30	1.84	1.90	1.90	1.95	2.90	2.95
STSP	-		105	290	3.48	3.30	3.00	3.00	2.78	2.48	1.78	1.60	1.48	1.30	1.30	1.30
SP-1	70	12	105	90	3.46	3.00	2.69	2.60	2.60	2.60	1.30	1.30	1.00	1.00	1.00	1.00
SP-2				135	3.00	2.48	1.69	1.60	1.30	1.30	<1	<1	<1	<1	<1	<1
SP-3				290	2.48	2.00	1.60	1.48	1.30	1.30	<1	<1	<1	<1	<1	<1
Raspberry purees																
USRP	70	12	-	-	3.45	3.60	3.60	3.95	4.00	4.30	1.90	2.00	2.30	2.30	2.90	3.00
STRP	-		105	290	3.44	3.30	2.95	2.84	2.84	2.60	1.84	1.60	1.60	1.30	1.00	1.00
RP-1	70	12	105	90	3.00	2.30	2.00	1.48	1.30	1.30	1.30	1.30	1.00	1.00	1.30	1.00
RP-2				135	2.47	2.30	1.69	1.30	1.30	1.00	<1	<1	<1	<1	<1	<1
RP-3				290	2.00	1.69	1.48	1.00	1.00	<1	<1	<1	<1	<1	<1	<1
Aronia purees																
USAP	70	12	-	-	3.48	3.47	3.60	3.95	3.95	4.00	1.69	1.84	2.00	2.00	2.48	3.00
STAP	-		105	290	3.30	2.90	2.78	2.30	2.30	2.00	1.60	1.30	1.30	1.00	1.00	1.00
AP-1	70	12	105	90	2.90	2.69	2.30	1.90	1.30	<1	<1	1.00	1.00	1.00	<1	<1
AP-2				135	2.30	1.90	1.30	1.00	<1	<1	<1	<1	<1	<1	<1	<1
AP-3				290	2.00	1.48	1.00	1.00	<1	<1	<1	<1	<1	<1	<1	<1

\* Note: USSP - ultrasonicated strawberry puree, STSP - sterilized strawberry puree, SP-1, SP-2, and SP-3 - strawberry puree sterilized for 90, 135, and 290 s, respectively; USRP - ultrasonicated raspberry puree, STRP - sterilized raspberry puree, RP-1, RP-2, and RP-3 - raspberry puree sterilized for 90, 135, and 290 s, respectively; USAP - ultrasonicated aronia puree, STAP - sterilized aronia puree, AP-1, AP-2, and AP-3 - aronia puree sterilized for 90, 135, and 290 s, respectively; TTU - thermal treatment by ultrasonication; TTS - thermal treatment by sterilization. Values represent the mean of three independent determinations ± standard deviation,  $P \leq 0.05$

## 4. STUDY OF THE INTERACTION BETWEEN GOAT - COW MILK MIXTURE WITH BERRY FRUIT PUREES IN YOGURT PRODUCTION

The main objective of the research presented in this chapter is to determine the optimal proportion between goat's milk and cow's milk for the production of natural yogurt, as well as to evaluate the quality indicators of yogurt made from a goat - cow milk mixture enriched with berry fruit purees during storage.

**4.1 Investigation of the fermentation process in the production of natural yogurts from a mixture of goat's and cow's milk.** To evaluate the influence of milk composition on the fermentation process, five samples of natural yogurts prepared from mixture of goat's and cow's milk were analyzed: S1, S2, S3, S4 and S5. The comparison of these formulations focused on the dynamics of acidification, expressed through pH evolution, and the accumulation of lactic acid as the main biochemical marker of fermentation. The evolution of pH values showed a progressive decrease across all samples, with similar final values but significant differences in the rate of acidification (Figure 6).



**Fig. 6. Evolution of pH during the fermentation of natural yogurts made from goat's and cow's milk mixture**

*\*Note: S1 - yogurt made from cow's milk*

*S2 - yogurt made from goat's milk*

*S3 - yogurt made from a goat's and cow's milk mixture in a 3:1 ratio*

*S4 - yogurt made from a goat's and cow's milk mixture in a 1:1 ratio*

*S5 - yogurt made from a goat's and cow's milk mixture in a 1:3 ratio*

The lactic acid content, analyzed at the same time intervals, followed a similar trend to the decrease in pH (Table 4). The highest lactic acid values at the end of fermentation (6 h) were observed in sample S4 (0.63 g/100 g), followed by S3 (0.57 g/100 g) and S5 (0.54 g/100 g), while samples S2 and S1 showed comparable levels (0.56 and 0.52 g/100 g, respectively). This hierarchy suggests that mixed formulations - particularly the balanced ones - more effectively stimulate the fermentative activity of the starter culture compared to milk from a single species. During the first two hours, all samples exhibited a slow accumulation of lactic acid, characteristic of the adaptation phase of lactic cultures. After four hours, a marked acceleration of the process was observed, indicating the onset of the logarithmic phase of bacterial multiplication.

**Table 4. Lactic acid content (g/100 g) accumulated during the fermentation of natural yogurts from goat and cow milk mixture**

No	Fermentation time, h	S1	S2	S3	S4	S5
1.	0	0.02±0.03	0.02±0.04	0.03±0.04	0.04±0.02	0.03±0.04
2.	2	0.11±0.06	0.12±0.03	0.12±0.05	0.14±0.07	0.13±0.04
3.	4	0.31±0.04	0.32±0.04	0.32±0.01	0.34±0.05	0.30±0.02
4.	6	0.52±0.02	0.56±0.02	0.57±0.02	0.63±0.02	0.54±0.02

*\*Note: S1 - yogurt made from cow's milk*

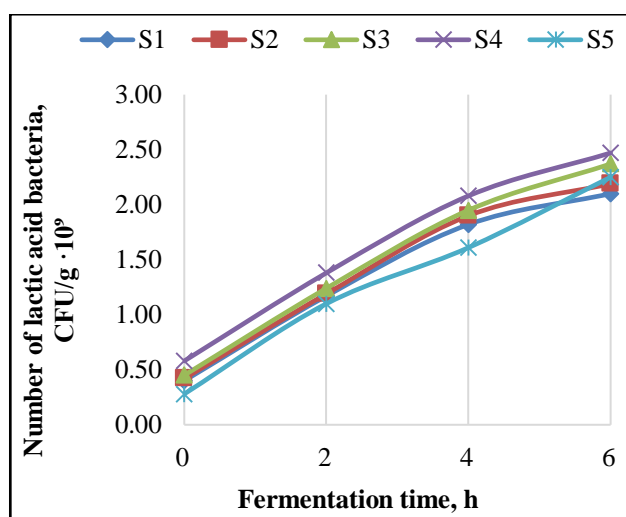
*S2 - yogurt made from goat's milk*

*S3 - yogurt made from a goat's and cow's milk mixture in a 3:1 ratio*

*S4 - yogurt made from a goat's and cow's milk mixture in a 1:1 ratio*

*S5 - yogurt made from a goat's and cow's milk mixture in a 1:3 ratio*

At the end of the fermentation process (6 h), a maximum accumulation of lactic acid was observed in all samples, indicating that the stationary phase had been reached and that the pH had stabilized around the optimal range for yogurt (4.5 - 4.7). During fermentation, a significant increase in the total number of lactic acid bacteria (LAB) was recorded across all samples, with variations depending on the milk composition. The results (Figure 7) highlight the influence of milk type on LAB growth, particularly during the first 2 hours of fermentation, when the metabolic activity of the starter culture is in the adaptation phase. At the beginning of fermentation, sample S4 exhibited the highest initial LAB concentration ( $0.58 \cdot 10^9$  CFU/g), suggesting an optimized environment for bacterial development. This value was higher compared to those recorded in the



**Fig. 7. Growth of lactic acid bacteria during the fermentation of natural yogurt from a goat and cow milk mixture**

*\*Note: S1 - yogurt made from cow's milk*

*S2 - yogurt made from goat's milk*

*S3 - yogurt made from a goat's and cow's milk mixture in a 3:1 ratio*

*S4 - yogurt made from a goat's and cow's milk mixture in a 1:1 ratio*

*S5 - yogurt made from a goat's and cow's milk mixture in a 1:3 ratio*

Values represent the mean of three independent determinations ± standard deviation,  $P \leq 0.05$

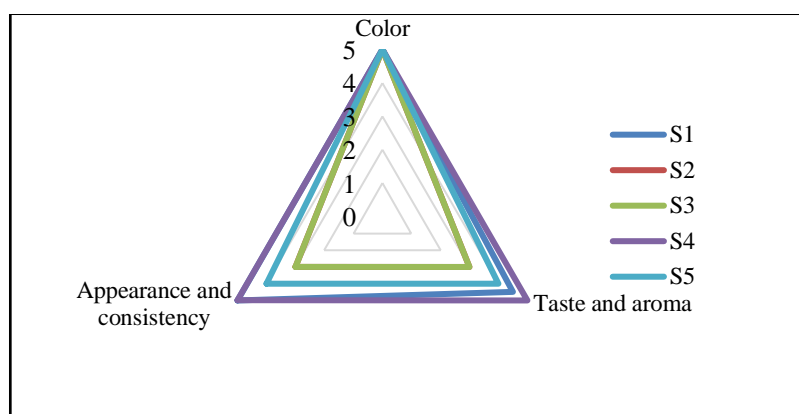
other samples: S3 -  $0.45 \cdot 10^9$  CFU/g, S5 -  $0.28 \cdot 10^9$  CFU/g, S2 -  $0.42 \cdot 10^9$  CFU/g, and S1 -  $0.40 \cdot 10^9$  CFU/g. These differences are attributed to the distinct chemical compositions of goat and cow milk, which influence the availability of essential nutrients for lactic acid bacteria multiplication, such as free amino acids, peptides, lactose, and minerals (calcium, phosphorus). During the 4 - 6 h interval, corresponding to the logarithmic phase of bacterial growth, a marked intensification of LAB multiplication was observed in all samples. The highest values were recorded in sample S4 ( $2.47 \cdot 10^9$  CFU/g), followed by

S3 ( $2.37 \cdot 10^9$  CFU/g), S5 ( $2.25 \cdot 10^9$  CFU/g), S2 ( $2.19 \cdot 10^9$  CFU/g), and S1 ( $2.10 \cdot 10^9$  CFU/g). This observation reconfirms that mixed formulations, particularly those balanced in a 1:1 ratio, create optimal conditions for the symbiotic relationship between *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus* - the lactic acid bacteria actively involved in yogurt fermentation. The synergistic activity between the two strains is essential for accelerating bacterial proliferation: *Lactobacillus bulgaricus*, through its proteolytic activity, releases peptides and amino acids that support the growth of *Streptococcus thermophilus*, while *Streptococcus thermophilus* produces metabolites such as carbon dioxide, formic acid, folic acid, pyruvic acid, and glutathione, which in turn stimulate the growth of *Lactobacillus bulgaricus*.

#### 4.2 Sensory evaluation of natural yogurts from a mixture of goat's and cow's milk.

The samples prepared from different goat and cow milk mixture (S3, S4, S5) exhibited intermediate behavior, with a semi - fluid consistency, homogeneous appearance, and variable reduction of syneresis. Among these, sample S4 stood out due to its optimal sensory balance: absence of syneresis, uniform white - yellow color, and a pleasant aroma characteristic of lactic fermentation, resulting in superior sensory evaluation.

Regarding aroma and taste, all samples exhibited profiles characteristic of lactic fermentation, without off - notes; however, the intensity of the aroma and the degree of acidity were decisive for the tasters' preferences. Samples with an optimal sugar - to - acid balance (notably S4 and S3) were well accepted, whereas excessive acidity or inappropriate texture (S2) resulted in lower scores. In this study, the sensory evaluation of natural yogurts from goat and cow milk mixture was conducted using a 5- point hedonic scale (Figure 8). All yogurt samples from



**Fig. 8. Sensory characteristics of natural yogurts made from goat and cow milk mixture**

\*Note: S1 - yogurt made from cow's milk

S2 - yogurt made from goat's milk

S3 - yogurt made from a goat's and cow's milk mixture in a 3:1 ratio

S4 - yogurt made from a goat's and cow's milk mixture in a 1:1 ratio

S5 - yogurt made from a goat's and cow's milk mixture in a 1:3 ratio

Values represent the mean of three independent determinations  $\pm$  standard deviation,  $P \leq 0.05$

the goat and cow milk mixture exhibited a characteristic aroma, described as pleasant, slightly tangy, and free from any off - odors or off - flavors, confirming the effective fermentative activity of the starter culture. However, samples S2 and S3 displayed a pronounced aftertaste of goat milk, which can be attributed to the presence of specific vo-

latile compounds and the distinctive fatty acid profile of this milk type, a phenomenon well documented in the specialized literature (Bulgaru et al., 2020). No unpleasant tastes such as bitterness, saltiness, or other off - flavors were detected in any of the samples, indicating correct fermentation and a well - controlled technological process. However, sample S2 recorded the lowest scores for taste (3.5 points) and aroma (3.0 points), which correlated with a weaker aroma and the presence of undesirable characteristics such as high whey separation, fluid consistency, and uneven appearance. These factors reduced overall acceptability, demonstrating that the composition of goat's milk can negatively affect the stability and sensory properties of yogurt. In contrast, sample S4 stood out for its superior sensory qualities, achieving the maximum score (5.0 points) due to a fine, homogeneous, and firm curd, absence of gas bubbles, and a balanced, pleasantly tangy taste without any off - flavors. These results indicate an optimal synergy in the 1:1 goat and cow milk mixture, allowing the combination of the nutritional benefits of both milk types.

**4.3 Physicochemical parameters of natural yogurts from a mixture of goat's and cow's milk.** The results obtained from the analysis of the physicochemical parameters of natural yogurts made from goat and cow milk mixture (Table 5) highlighted significant features related to product composition, primarily influenced by the type and proportion of milk used in the formulation. Fat content was highest in sample S1 (4.41%), made exclusively from cow's milk, which can be explained by the intrinsic differences between goat and cow milk compositions. Total protein content varied slightly among the natural yogurt samples from goat and cow milk mixture, with the highest values observed in samples S2 (5.36%) and S1 (5.34%).

**Table 5. Physicochemical parameters of natural yogurts made from a mixture of goat's and cow's milk**

No	Parameters	S1	S2	S3	S4	S5
1.	Fat content, %	4.41±0.04	3.10±0.07	3.5±0.05	3.4±0.05	3.6±0.04
2.	Total protein content, %	5.34±0.03	5.36±0.04	5.25±0.05	5.31±0.04	5.28±0.06
3.	Water activity, c.u.	0.883±0.002	0.884±0.001	0.880±0.002	0.881±0.002	0.882±0.001
4.	Ash content, %	0.72±0.13	0.75±0.16	0.74±0.12	0.76±0.15	0.73±0.14
5.	Total solids content, %	18.13±0.23	17.94±0.25	17.38±0.28	17.80±0.30	17.28±0.31
6.	Non-fat dry matter content, %	14.34±0.26	14.24±0.25	13.98±0.22	14.30±0.23	14.11±0.21
7.	Milk pasteurization control - phosphatase	absence	absence	absence	absence	absence

*\*Note: S1 - yogurt made from cow's milk*

*S2 - yogurt made from goat's milk*

*S3 - yogurt made from a goat's and cow's milk mixture in a 3:1 ratio*

*S4 - yogurt made from a goat's and cow's milk mixture in a 1:1 ratio*

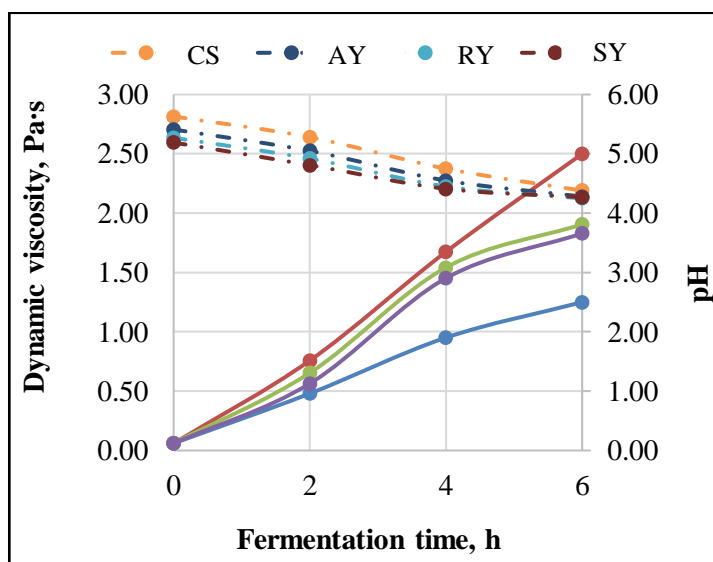
*S5 - yogurt made from a goat's and cow's milk mixture in a 1:3 ratio*

*Values represent the mean of three independent determinations ± standard deviation, P≤0.05*

The water activity ( $a_w$ ) values ranged between 0.880 and 0.884 c.u., indicating a moderate availability of water for microorganisms. The ash content was slightly higher in the natural yogurt samples containing goat milk (S2, S3, S4), with the following results: S1 - 0.72%, S2 - 0.75%, S3 - 0.74%, S4 - 0.76%, and S5 - 0.73%. The total solids content showed the following values: S1 - 18.13%, S2 - 17.94%, S3 - 17.38%, S4 - 17.80%, and S5 - 17.28%. The non - fat dry matter content was as follows: S1 - 14.24%, S2 - 14.24%, S4 - 14.30%, S3 - 13.98%, and S5 - 14.11%. The observed variations in the physicochemical indicators of natural yogurts made from goat and cow milk mixtures are directly correlated with the type and proportion of milk used, influencing sensory quality, microbiological stability, and nutritional value.

#### 4.4 Development of yogurts from a mixture of goat's and cow's milk with berry fruit purees

**4.4.1 Correlation between dynamic viscosity and pH during fermentation of yogurts from a mixture of goat's and cow's milk with berry fruit purees.** The evolution of DV in yogurts with berry purees compared to the sample CS was monitored throughout the 6- hour fermentation period (Figure 9). During the first 2 hours of fermentation, changes in DV were minimal, followed by a progressive increase between 2 and 4 hours, reaching maximum values at the end of fermentation (6 h). The DV ranges for the yogurts from the goat and cow milk mixture with berry purees were as follows: sample AY (with aronia): 0.06 - 2.50 Pa·s, sample RY (with raspberry): 0.06 - 1.54 Pa·s, and sample SY (with strawberry): 0.06 - 1.45 Pa·s, compared to the sample CS: 0.06 - 1.25 Pa·s. The highest value was recorded in sample IA, which can be attributed



**Fig. 9. The evolution of dynamic viscosity and pH during the fermentation of yogurts made from a mixture of goat and cow milk with berry fruit purees**

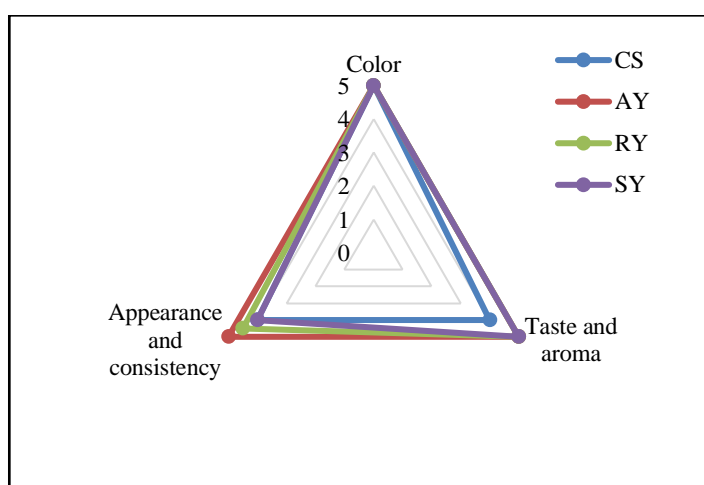
*\*Note: CS - control sample; AY - aronia yogurt; RY - raspberry yogurt; SY - strawberry yogurt*

to the higher content of soluble fibers and pectin in the aronia puree, promoting the formation of a denser and more stable gel network. The increase in DV was closely correlated with the decrease in pH during fermentation. The pH reduction, driven by the activity of LAB, promoted the formation of a semi-solid three-dimensional gel structure in the yogurt by facilitating casein micelle aggregation and strengthening intermolecular bonds within the yogurt matrix.



These changes were accompanied by increased water retention and reduced syneresis, contributing to improved viscosity and firmness of the final product. Additionally, the dietary fiber and polysaccharide content- particularly pectin - derived from the berry purees positively influenced the gel structure.

**4.4.2 Sensory evaluation of yogurts from a mixture of goat's and cow's milk with berry fruit purees.** Based on the scores obtained (Figure 10), all yogurts made from a mixture of goat's and cow's milk with berry purees received high evaluations for sensory characteristics, with scores ranging from 3.5 to 5.0 points for each attribute assessed. The most highly rated attributes were taste and aroma, followed by appearance and consistency. In particular, sample AY recorded the highest overall acceptability scores, featuring a smooth and creamy texture, a well - balanced



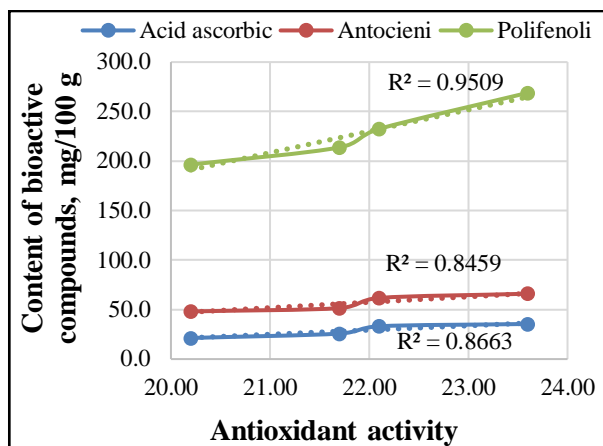
**Fig. 10 Sensory characteristics of yogurts made from a mixture of goat's and cow's milk with berry purees**

*\*Note: CS - control sample; AY - aronia yogurt; RY - raspberry yogurt; SY - strawberry yogurt*

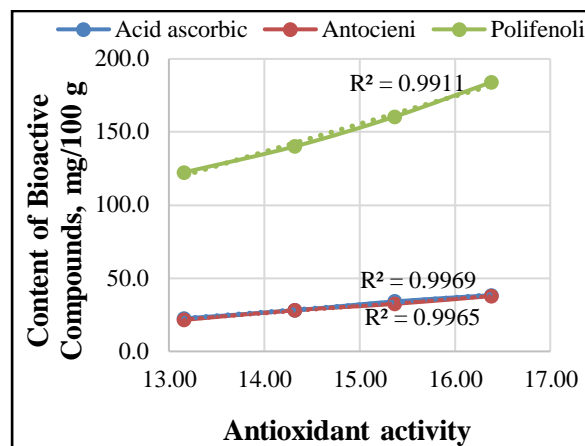
sweet - tart taste, an attractive pale-violet color, and a pleasant, natural aroma. These results indicate that berry purees not only enhanced the sensory profile of the goat - cow milk yogurt but also increased its overall acceptability, with no defects detected by the evaluators during sensory analysis. Thus, berry purees can be considered functional ingredients in the formulation of goat - cow milk yogurts enriched with berry purees.

**4.5 Effect of storage time on the antioxidant activity and biological value of yogurts from a mixture of goat's and cow's milk with berry purees.** The correlation between antioxidant activity and the content of *L*- ascorbic acid, total anthocyanins, and polyphenols was assessed throughout the storage period of the yogurts by calculating the coefficient of determination ( $R^2$ ) (Figure 11). The results revealed significant differences among the yogurts containing berry purees. In sample AY, the strongest correlation was observed between antioxidant activity and the content of polyphenols and anthocyanins, whereas *L*- ascorbic acid exhibited a moderate coefficient of determination ( $R^2 = 0.8663$ -  $0.9969$ ), indicating a comparatively lower contribution to the overall free radical scavenging capacity. In sample RY, the total polyphenol content showed the strongest correlation with antioxidant activity ( $R^2 = 0.9509$  -  $0.9983$ ), while both total anthocyanin content and *L*- ascorbic acid also exhibited high and comparable values ( $R^2$

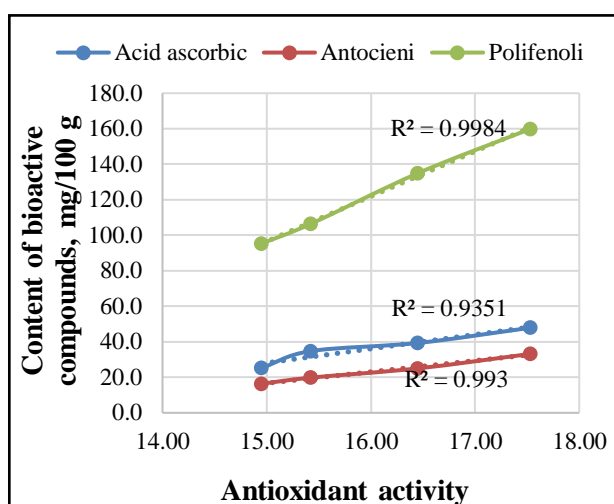
= 0.8459 - 0.9969 and 0.8663 - 0.9969, respectively), confirming their similar role in maintaining antioxidant stability. For sample SY, total polyphenols remained the primary determinants of antioxidant activity ( $R^2 = 0.9509 - 0.9984$ ); however, *L* -ascorbic acid demonstrated a stronger correlation than total anthocyanins, suggesting a specific characteristic of this food matrix in which



a) AY



b) RY



**Fig. 11 The correlation between antioxidant activity and the biological value of goat - cow milk mixture yogurts with berry purees during storage**

c) SY

*\*Note: AY - aronia yogurt; RY - raspberry yogurt; SY - strawberry yogurt*

*L*-ascorbic acid plays a more significant role in overall antioxidant capacity. Correlation analysis indicated that total polyphenols ( $R^2 = 0.92$ ,  $p < 0.01$ ) and anthocyanins ( $R^2 = 0.86$ ,  $p < 0.01$ ) contribute substantially to total antioxidant activity, while *L*-ascorbic acid has a more modest, yet still relevant, influence ( $R^2 = 0.85$ ,  $p < 0.05$ ). Consequently, the regression analysis and the high  $R^2$  values confirm that the interaction of bioactive compounds is a key factor determining antioxidant activity during the storage of berry puree yogurts.

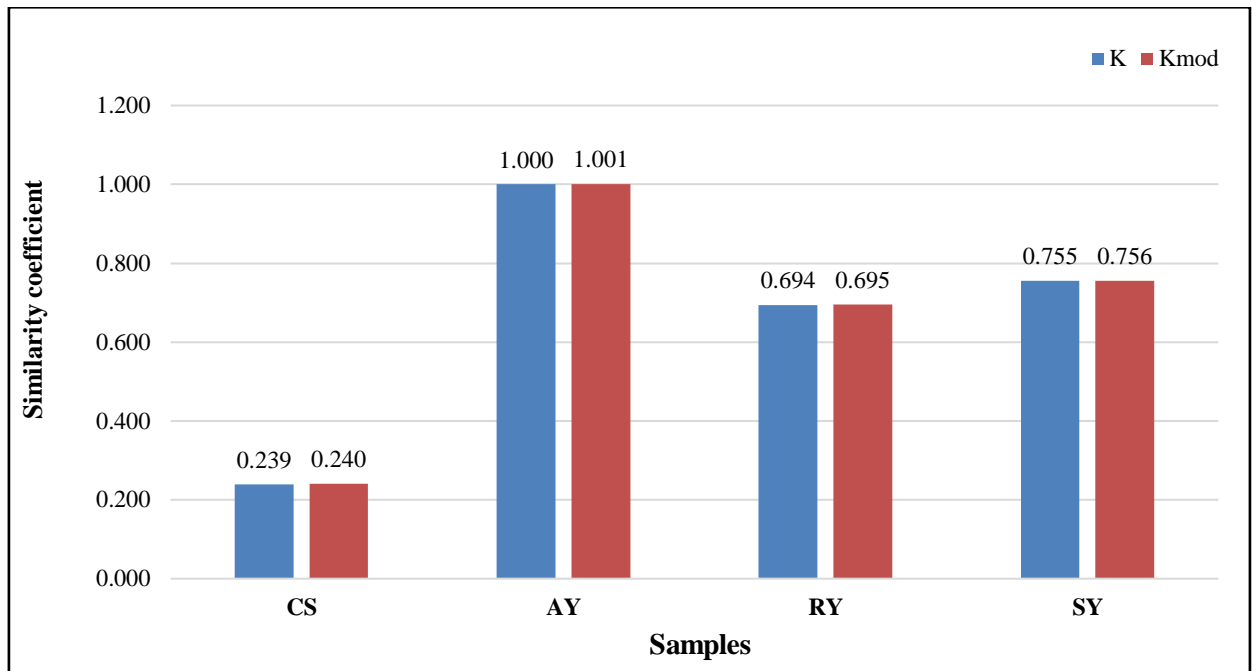
**4.6 Mathematical modeling of the final yogurts from a mixture of goat's and cow's milk with berry fruit purees.** The mathematical model describing the quality of yogurts produced from goat and cow milk mixture enriched with berry purees demonstrated a strong correlation of approximately 98.25% with the results obtained from analyses of sensory characteristics, microbiological and physicochemical indicators, biological value, and antioxidant activity. The

mathematical model, based on statistical data collected during passive experimentation and developed using correlation and regression analysis methods, was formulated as follows:

$$K = 1.001 \cdot (-9,0783pH^2 + 97,29pH - 259,73) \cdot (-16,188LA^3 + 141,81LA^2 - 411,01LA + 395,24) \cdot (-0,0001TA + 0,0052) \cdot (-13,249PC^2 + 4,1634FC + 0,6593) \cdot (0,1284DV^3 - 0,7108DV^2 + 1,2584DV + 0,2911) \cdot (0,0004LAB + 1) \cdot (-2 \cdot 10^{-5}TSC + 1.0003) \cdot (5 \cdot 10^{-7}AA + 1) \cdot (-3 \cdot 10^{-6}AC + 1) \cdot (4 \cdot 10^{-8}a_w + 1) \cdot (-5 \cdot 10^{-12}P^3 + 5 \cdot 10^{-9}P - 10^{-6}P + 1) \cdot (4 \cdot 10^{-7}AA + 1) \cdot (10^{-5}P + 0,999)$$

*\*Note: LA- lactic acid, TA- titratable acidity, PC- protein content, FC- fat content, LAB- lactic acid bacteria, DV- dynamic viscosity, TSC - total solids content, AA- antioxidant activity, AC- ash content,  $a_w$ - water activity*

Figure 12 illustrates the similarity between the values predicted by the model and the experimental data for the yogurts made from mixture goat's and cow's milk with berry purees. The high reliability of the model is further supported by the Fisher statistical criterion, for which the calculated value exceeded the minimum required threshold of 1.00. This indicates that the model is statistically significant and that variations in the input parameters account for a substantial proportion of the variability in the output parameters.



**Fig. 12 Similarity of the mathematical model of the quality of goat's and cow's milk mixture yogurts with berry fruit purees**

*\*Note: CS- control yogurt, AY - aronia yogurt; RY - raspberry yogurt; SY - strawberry yogurt*

Therefore, the proposed mathematical model proved to be an adaptable and reliable tool for evaluating and optimizing the quality of goat's and cow's milk mixture yogurts with berry fruit

purees, providing a solid foundation for process control and for adjusting manufacturing parameters aimed at improving product quality.

## **GENERAL CONCLUSIONS AND RECOMMENDATIONS**

The study and analysis of the results obtained within the doctoral thesis in engineering sciences entitled “IMPROVEMENT OF THE BIOLOGICAL VALUE OF FERMENTED DAIRY PRODUCTS WITH BIOACTIVE INGREDIENTS FROM FRUITS” led to the formulation of the following conclusions:

1. The application of ultrasonic thermal treatment (60 - 70 °C for 5 - 15 min) contributed to obtaining high - quality berry fruit purees; however, its microbiological stability remained limited (5.95 log CFU/g) due to the persistence of thermoresistant yeasts. In contrast, conventional thermal sterilization (105 °C for 90 - 290 s) ensured complete inactivation of both vegetative microflora and spores, guaranteeing the microbiological safety of berry purees. Therefore, combining ultrasonic preservation with thermal sterilization proved to be an optimal technological solution, allowing the production of microbiologically safe (3.00 log CFU/g) and biologically functional purees, with the following values: *L*- ascorbic acid - 72.37 mg/100 g, total anthocyanins content - 150.51 mg/100 g, antioxidant activity - 10.28 mg TE/100 g (subchapter 3.1).

2. The combined thermal treatment consisting of ultrasonication at  $70 \pm 1$  °C for  $12 \pm 2$  min, followed by sterilization at 105 °C for 135 s, was identified as the optimized preservation regime for berry purees. This strategy ensured the retention of bioactive compounds, including up to 78.43% of total anthocyanins content and 63.65% of *L*- ascorbic acid, while maintaining 53.66% of antioxidant activity and significantly reducing pathogenic and spoilage microorganisms by 94.23%. Too short a duration favored microbial persistence, where as excessive processing caused marked degradation of bioactive compounds, underlining the need for precise calibration of the combined treatment (Cuşmenco et al., 2024; subchapter 3.3).

3. The development of yogurt made from a 1:1 mixture of goat's and cow's milk demonstrated that this formulation optimized the fermentation process and resulted in a yogurt with superior sensory qualities - creamy consistency, balanced taste, and uniform white-yellowish color. The resulting environment was unfavorable to spoilage and pathogenic microorganisms. The physicochemical parameters were within the following ranges: pH 4.53 - 4.60, lactic acid 0.54 - 0.63 g/100 g, *LAB*  $2.25 - 2.47 \cdot 10^9$  CFU/g, fat content 3.1 - 3.5%, total protein content 5.25 - 5.36 g/100 g, water activity 0.881 - 0.884 c.u., ash content 0.73 - 0.76%, and total solids content 17.28 - 17.94% (Bulgaru et al., 2020; Cuşmenco et al., 2022; subchapter 4.1).

4. Yogurts made from the goat - cow milk mixture with berry fruit purees showed improved physicochemical and microbiological indicators, as follows: titratable acidity 85.74 - 91.35 °T, pH

4.25 - 4.38, water activity 0.858 - 0.871 c.u., total solids content 18.11 - 18.45%, ash content 0.69 - 0.89%, total protein content 5.23 - 5.87%, lactic acid 0.69 - 0.89 g/100 g, and dynamic viscosity 1.45 - 2.50 Pa·s. Regarding the biological value, the levels ranged from 35.8 - 47.7 mg/100 g for *L*-ascorbic acid, 33.1 - 66.3 mg/100 g for total anthocyanins content and 159 - 268 mg GAE/100 g for total polyphenols content. The antioxidant activity ranged from 13.5 to 20.2 mg TE/100 g (Cuşmenco et al., 2021; subchapter 4.2).

5. Statistical analysis and mathematical modeling of the experimental data revealed strong correlations between bioactive compounds and the antioxidant activity of goat - cow milk mixture yogurts with berry purees. The multiple linear regression model demonstrated that polyphenols and anthocyanins were the main contributors to antioxidant stability, with determination coefficients  $R^2 = 0.9984$  and  $R^2 = 0.9969$  ( $p < 0.01$ ), while *L*-ascorbic acid exerted a moderate but significant influence ( $R^2 = 0.8663$ ,  $p < 0.05$ ). The developed mathematical model exhibited a correlation of approximately 98.25% with experimental results and met Fisher's statistical criterion (0.695 - 1.01), confirming both its reliability and validity for describing and predicting technological processes (subchapters 4.11 and 4.13).

## PRACTICAL RECOMMENDATIONS

- ❖ It is proposed to apply the combined thermal treatment, which integrates ultrasonication and thermal sterilization, as the optimal preservation method for fruit purees made from aronia, raspberry, and strawberry. The optimized parameters for ultrasonication are: temperature  $70 \pm 1^\circ\text{C}$ , exposure time  $12 \pm 2$  min, frequency 37 kHz, and power 510 W; for sterilization –  $105^\circ\text{C}$  for 135 s. This combined protocol ensures the effective preservation of biologically active compounds and the maintenance of antioxidant activity, while achieving a significant reduction of the total preservation time by approximately 53.44%.
- ❖ Based on the results obtained, it is recommended to use an optimal mixture of goat and cow milk in a 1:1 (v/v) ratio with berry fruit purees. This formulation has proven favorable for the development of yogurt with improved rheological properties. The proposed formula enhances the compositional synergy between the two types of milk, ensuring optimal structural stability and high sensory acceptability.
- ❖ The yogurt production technology from goat and cow milk should be implemented according to the invention patent.

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## ABSTRACT

**Cuşmenco Tatiana: “Improvement of the biological value of fermented dairy products with bioactive fruit ingredients,” PhD thesis in engineering sciences, Chişinău, 2025.**

**Structure of the thesis:** The thesis includes an introduction, 4 chapters, conclusions and recommendations, and a bibliography containing 260 references. The main body of the text comprises 139 pages, including 49 figures and 35 tables. The results obtained have been published in 19 scientific papers.

**Keywords:** goat milk, cow milk, fruits, starter culture, yogurt, technology, fermentation, acidity, lactic acid bacteria, protein, viscosity, texture, firmness, syneresis, antioxidants, stability.

**The purpose of the work:** obtain berry fruit purees preserved through a combined method of ultrasonication and sterilization, ensuring high antioxidant potential, and to use these purees in the production of yogurt with enhanced biological value.

**The objectives of the paper:** investigate the influence of ultrasonication and sterilization temperature on the biological value and high antioxidant potential of berry fruit purees; establish mathematical models for optimizing the preservation conditions of berry fruit purees using kinetic modeling; determine the influence of ultrasonication and sterilization conditions on physicochemical parameters (titratable acidity, pH, total solids, ash, protein, fiber, sugars), biological value (TAC, *L*- ascorbic acid), and antioxidant activity (AA) of the purees, and to define optimal preservation conditions; determine the optimal ratio of goat - cow milk mixture for natural yogurt production; develop a manufacturing technology for yogurt obtained from goat - cow milk mixtures; evaluate the effect of storage duration on the sensory, microbiological, physicochemical qualities, and antioxidant potential of yogurts with berry purees; apply mathematical modeling for evaluating the quality of fruit yogurt as a final product.

**Scientific novelty and originality:** for the first time, the optimal preservation conditions for berry fruit purees were determined and identified using a combined thermal treatment of ultrasonication and sterilization, which enables the retention of biological value and antioxidant activity. The importance of using a goat - cow milk mixture in the production of fermented dairy products was substantiated, and the necessity of enriching yogurt made from goat - cow milk mixture with berry was justified in terms of biologically active compound content and microbiological stability.

**Main results:** the combined effect of ultrasonication and sterilization on the biological value and antioxidant potential was analyzed; the optimization of thermal treatment parameters and their effect on microbiological stability, physicochemical indices, sensory characteristics, bioactive compounds, and antioxidant activity of the purees was substantiated; the influence of these factors on the fermentation process and yogurt quality parameters was demonstrated; the stability of quality indicators in fruit yogurts during storage was also confirmed.

**Theoretical significance:** for the first time, kinetic modeling of the preservation method using thermal treatment by ultrasonication and sterilization of berry purees under various temperature and duration conditions has been performed; optimal conditions for preservation through thermal treatment were determined, focusing on their impact on physicochemical characteristics, biological value, and antioxidant activity; technologies for yogurt production using berry purees as bioactive filling agents were developed to enhance the final product's biological value.

**Applicative value:** a yogurt production process has been developed, resulting in invention patent and implementation act of the industrial - scale yogurt manufacturing technology with fruits at the specialized company “Ferma cu Origini” LDT.



## ADNOTARE

**Cuşmenco Tatiana:** „Ameliorarea valorii biologice a produselor lactate fermentate cu ingrediente bioactive din fructe”, teză de doctor în științe ingineresti, Chișinău, 2025.

**Structura tezei:** constă din introducere, 4 capitole, concluzii și recomandări, bibliografie cu 260 titluri. Textul de bază conține 139 pagini, inclusiv 49 de figuri și 35 de tabele. Rezultatele obținute sunt publicate în 19 lucrări științifice

**Cuvinte-cheie:** lapte de capră, lapte de vacă, fructe, cultură starter, iaurt, tehnologie, fermentare, aciditate, bacterii lactice, proteină, vâscozitate, textură, fermitate, sinereză, antioxidanți, stabilitate.

**Scopul lucrării:** constă în obținerea piureurilor din fructe bacifere conservate prin metoda combinată de ultrasonificare și sterilizare cu păstrarea potențialului antioxidant ridicat și utilizarea acestora în fabricarea iaurtului cu valoare biologică ameliorată.

**Obiectivele lucrării:** cercetarea influenței condițiilor de ultrasonificare și sterilizare asupra valorii biologice și a potențialului antioxidant a piureurilor din fructe bacifere și stabilirea modelelor matematice pentru determinarea condițiilor optime de conservare; determinarea condițiilor optime de ultrasonificare și sterilizare asupra caracteristicilor senzoriale, indicatorilor fizico - chimici (aciditatea titrabilă, pH, conținut de substanță uscată, cenușă, proteină, fibre, zahăr), valorii biologice (acid *L*- hidroascorbic, conținutului total de antocieni) și activității antioxidante a piureurilor din fructe bacifere; determinarea raportului optim pentru elaborarea tehnologiei de fabricare a iaurtului din amestec de lapte de capră și vacă; cercetarea influenței piureului din fructe bacifere în iaurt și duratei de păstrare a acestuia asupra caracteristicilor senzoriale, indicatorilor fizico - chimici, stabilității microbiologice, valorii biologice, activității antioxidante și stabilirea modelului matematic privind calitatea produsului finit.

**Noutatea și originalitatea științifică:** pentru prima dată au fost determinate și identificate condițiile optime de conservare a piureurilor din fructe bacifere prin tratament termic combinat de ultrasonificare și sterilizare care permite păstrarea valorii biologice și activității antioxidante; a fost argumentată importanța utilizării amestecului din lapte de capră și vacă în fabricarea produselor lactate fermentate; a fost fundamentată necesitatea îmbogățirii iaurtului din amestec de lapte de capră și vacă cu fructe prin prisma conținutului de compuși biologic activi și stabilității microbiologice.

**Rezultatele principale:** s-a stabilit influența tratamentului termic combinat prin ultrasonificare și sterilizare asupra valorii biologice și potențialului antioxidant ale piureurilor din fructe bacifere; s-au determinat condițiile optime de ultrasonificare și sterilizare care asigură conservarea caracteristicilor senzoriale, a indicatorilor fizico - chimici, precum și a compușilor biologic activi; s-a stabilit raportul optim dintre laptele de capră și laptele de vacă pentru elaborarea tehnologiei de fabricare a iaurtului, în vederea obținerii unui produs cu proprietăți funcționale îmbunătățite și o valoare biologică ridicată; s-a demonstrat influența adaosului de piureuri din fructe bacifere și a duratei de păstrare asupra iaurturilor obținute, evidențiindu-se menținerea caracteristicilor senzoriale, a stabilității microbiologice, a indicatorilor fizico - chimici și a compușilor bioactivi pe perioada de păstrare. În plus, s-au elaborat modele matematice care descriu evoluția calității produsului finit și care permit estimarea stabilității acestuia în condiții de păstrare.

**Semnificația teoretică:** pentru prima dată a fost realizată modelarea cineticii procesului de conservare prin tratament termic de ultrasonificare și sterilizare a piureurilor din fructe bacifere, în funcție de temperatură și durată; au fost fundamentate particularitățile influenței tratamentului combinat prin ultrasonificare și sterilizare, fiind evidențiate corelațiile dintre indicatorii fizico - chimici, valoarea biologică și activitatea antioxidantă; a fost elaborată, în care piureurile din fructe bacifere au fost utilizate ca umplutură funcțională, contribuind la îmbogățirea valorii biologice și la creșterea stabilității produsului finit.

**Valoarea aplicativă:** a fost realizat procedeu de obținere a iaurtului din amestec de lapte de capră și vacă cu piureuri din fructe bacifere, a fost obținut brevet de invenție și act de implementare în cadrul întreprinderii de profil SRL „Ferma cu Origini”.

## АННОТАЦИЯ

**Кушменко Татьяна:** « Улучшение биологической ценности кисломолочных продуктов с использованием биоактивных фруктовых ингредиентов», диссертация на соискание ученой степени доктора инженерных наук, Кишинэу, 2025.

**Структура диссертации:** состоит из введения, 4 глав, выводов и рекомендаций, библиография в 260 наименованиях. Основной текст содержит 139 страниц, в том числе 49 рисунков и 35 таблицы. Полученные результаты опубликованы в 19 научных статьях.

**Ключевые слова:** козье молоко, коровье молоко, фрукты, стартовая культура, йогурт, технология, ферментация, кислотность, молочнокислые бактерии, белок, вязкость, текстура, синерезис, антиоксиданты, стабильность.

**Цель работы:** получение фруктовых пюре из ягод, консервированных с использованием комбинированного метода ультразвуковой обработки и стерилизации при сохранении высокого антиоксидантного потенциала, а также использование этих пюре при производстве йогурта с повышенной биологической ценностью.

**Задачи работы:** изучение влияния температуры ультразвуковой обработки и стерилизации на биологическую ценность и антиоксидантный потенциал фруктовых пюре из ягод и разработка математических моделей оптимальных условий на основе кинетических моделей; определение влияния условий ультразвуковой обработки и стерилизации на физико - химические параметры, биологическую ценность и антиоксидантную активность фруктовых пюре; определение оптимальных соотношений смеси козьего и коровьего молока для производства йогурта; изучение влияния срока хранения на органолептические, микробиологические, физико - химические характеристики и антиоксидантный потенциал йогурта с фруктовыми пюре; постановка математического моделирования качества йогурта с фруктовыми пюре конечного продукта.

**Научная новизна и оригинальность:** впервые были определены и идентифицированы оптимальные условия консервирования фруктовых пюре с использованием комбинированной термической обработки - ультразвука и стерилизации, что обеспечивает сохранение биологической ценности и антиоксидантной активности; обоснована важность использования смеси козьего - коровьего молока при производстве йогурта и необходимость обогащения йогурта с фруктовыми пюре с точки зрения содержания биологически активных веществ и микробиологической стабильности.

**Основные результаты:** проанализировано влияние комбинированной термической обработки на биологическую ценность и антиоксидантный потенциал фруктовых пюре; обосновано влияние оптимизации параметров обработки на микробиологическую стабильность, физико - химические показатели, органолептические характеристики и антиоксидантную активность; продемонстрировано влияние этих факторов на процесс ферментации и показатели качества йогурта; доказана стабильность качественных показателей фруктовых йогуртов в течение хранения.

**Теоретическая значимость:** впервые выполнено кинетическое моделирование процесса консервирования фруктовых пюре с использованием термической обработки ультразвуком и стерилизацией при различных температурных и временных режимах; установлены оптимальные условия для сохранения физико - химических показателей, биологической ценности и антиоксидантной активности; разработаны технологии производства йогуртов с использованием фруктовых пюре в качестве наполнителя с целью обогащения биологической ценности конечного продукта.

**Прикладное значение:** разработан технологический процесс производства йогурта; получен патент на изобретение и оформлен акт внедрения технологии промышленного производства фруктового йогурта на профильном предприятии АО „Ferma cu Origini”.

**CUȘMENCO TATIANA**

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**Summary of the doctoral thesis in engineering sciences**

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