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Evaluation of technological parameters in the production of beverages based on fermented plant ingredients

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Abstract. The aim of the study was to determine the optimal conditions for the fermentation of plant raw materials to ensure the stability and a favourable combination of sensory, microbiological, and physicochemical properties in beverages. The research was conducted under laboratory conditions and involved the fermentation of beverages from plant-based ingredients using both traditional and combined technologies. Physicochemical (pH, titratable acidity, redox potential), microbiological (counts of lactic acid bacteria and yeasts), and sensory parameters

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were monitored. It was found that combined fermentation resulted in a more rapid reduction in beverage pH (down to 4.0 within 72 hours) compared to traditional lactic acid fermentation (pH 4.6). The lowest pH (3.8) and highest titratable acidity (8.5 g/L) were observed in beverages based on fruit extracts, contributing to enhanced microbiological stability. Kombucha-based beverages exhibited intermediate acidity (6.5 g/L), while those produced from cereal substrates had the lowest acidity (5.7-6.0 g/L) and the shortest shelf life (10-12 days). Sensory analysis confirmed the advantages of combined fermentation: beverages produced using this method achieved a more complex aromatic profile and balanced taste, scoring 8.3 on a nine-point scale. Microbiological analysis revealed that yeast populations peaked at 48 hours (7.8 log CFU/mL) before declining, whereas lactic acid bacteria continued to increase until the end of fermentation (7.5-7.1 log CFU/mL). Correlation analysis confirmed the influence of acidity on both microbial composition and sensory characteristics. The results demonstrated that combined fermentation leads to a more significant pH reduction, increased titratable acidity, and stabilisation of redox potential, thereby improving the microbiological stability and sensory quality of beverages. This approach enables the optimisation of functional fermented beverage production, enhancing organoleptic properties and extending shelf life

Keywords: fermentation; acidity; redox potential; lactic acid bacteria; yeast; sensory analysis; microbiological stability

Introduction

Fermented beverages based on plant-derived raw materials are gaining increasing popularity due to their functional properties, high nutritional value, and potential health benefits. The fermentation process enhances the bioavailability of nutrients, reduces the content of anti-nutritional compounds, and contributes to the development of a distinctive sensory profile. In particular, lactic acid and yeast fermentation promote the synthesis of bioactive compounds – such as organic acids, poalyphenols, and B vitamins – which increase the antioxidant activity and probiotic potential of the beverages. However, the efficiency of the fermentation process largely depends on the selection of microbial cultures, fermentation parameters, and the type of plant raw material used (Dudarev, 2024). These factors underline the importance of optimising technological regimes to produce a high-quality and stable final product. Despite growing interest in plant-based fermented beverages, the control of physicochemical parameters during production, as well as the influence of different microbial strains on beverage quality, remains insufficiently studied.

Globally, the development of innovative fermentation methods for plant-based beverages is recognised as a priority area within the broader strategies for healthy diets and food security (Bekbayev *et al.*, 2024). The World Health Organization (n.d.) and the Food and Agriculture Organization (n.d.) highlight the role of fermented foods in enhancing the nutritional quality of diets and reducing the risk of chronic diseases (Ismayilov *et al.*, 2023). Numerous studies have examined the impact of technological parameters on the physicochemical properties, microbiological stability, and organoleptic characteristics of fermented plant-based beverages. Research has focused on modern processing methods, safety control, and fermentation optimisation. For instance, the study by M. Karputina & S. Oliinyk (2024) demonstrated that optimising fermentation processes improves the quality and microbiological safety of low-alcohol plant-based beverages, particularly through the selection of specific microbial strains and adjustment of technological conditions. R. Hrushchetskyi *et al.* (2023) highlighted the potential of Ukrainian plant raw materials – including rare vegetables and fruits – for creating innovative fermented beverages with both high nutritional value and distinctive sensory attributes. The study by K. Kondratenko (2023) focused on the development of fermented beverage technologies from vegetable substrates, showing that fermentation time and the choice of starter culture significantly influence acidity, stability, and consumer acceptance of the final product. Meanwhile, C. Penha *et al.* (2021) investigated the use of environmentally friendly technologies in producing plant-based beverages, particularly fermentation using probiotic cultures, which contributed to preserving sensory qualities and enhancing nutritional value.

Considerable attention has been devoted to enhancing the functional properties of fermented beverages through the optimisation of fermentation processes and the incorporation of additional bioactive components. A. Keşa *et al.* (2021) investigated strategies to improve the biological activity of fruit-based beverages by employing probiotic cultures and regulating fermentation parameters. Similarly, S. Liang *et al.* (2021) analysed technological processes in the production of tea-based beverages, focusing on the integration of traditional and innovative methods to enhance both sensory and chemical characteristics. L. Zhang *et al.* (2021) reviewed recent technological advancements aimed at eliminating defects in the production of fermented plant products, particularly the use of alternative fermentation methods to preserve nutritional value. Significant emphasis on biotechnological approaches to the production of fermented beverages from non-traditional plant raw materials was evident in the study by A. Vavilova (2023), which underscored the influence of microorganism type and technological conditions on the stability, chemical composition, and sensory acceptability of the final product. O. Kovalova *et al.* (2024a) note that "the use of plasma-chemically activated aqueous solutions increases the efficiency of enzymatic processing of lentils, improves the quality of the malted product, and promotes the activation of biochemical processes important for the production of functional beverages". In a related study, O. Kovalova *et al.* (2024b) emphasise that "the use of plasma-chemically activated water in the production of oat malt allows for the achievement of stable acidity indicators and the optimisation of enzymatic activity in the substrate, which is a prerequisite for high-quality fermentation". I. Ivanova *et al.* (2024) demonstrate that "the use of ridge regression to predict acidity and dry matter content in fruits enables the effective selection of raw materials with suitable fermentation characteristics – an essential factor in ensuring beverage stability and flavour".

Despite advances in research, the relationships between fermentation parameters, microbiological changes, and organoleptic characteristics – as well as the effects of different combinations of microorganisms on beverage stability and functional properties – remain insufficiently explored. The aim of this study was to evaluate the technological parameters of fermentation and their impact on the quality of beverages derived from plant-based raw materials. The specific objectives were to determine the influence of enzymatic agents and fermentation parameters on acidity, redox potential, and organoleptic characteristics; to analyse microbiological changes and their correlation with the physicochemical properties of the product; and to assess beverage stability depending on the choice of raw materials and fermentation conditions.

Materials and Methods

The experimental work was conducted from April to November 2024 at the Department of Food Biotechnology and the Laboratory of

Microbiological Quality Control of the National University of Food Technologies (NUFT) in Kyiv, Ukraine. Physicochemical analyses – including the determination of redox potential, acidity, and sugar content – were performed at the Central Research Laboratory of NUFT using certified equipment in accordance with Good Laboratory Practice (GLP) standards (European Food Safety Authority, n.d.). Seven groups of plant-based raw materials, differing in biochemical composition, were selected for fermentation: oats, millet, and barley (cereals); pomegranate, apple, and citrus extracts (fruits); and kombucha infusion (tea mushroom). Oat grain of the Harmony variety (Limited Liability Company "Agroprom-Lan", Kyiv region) was cleaned, washed, dried, crushed to a particle size of 0.8-1.2 mm, and extracted at 95°C for 30 minutes. Millet (Yavir) and barley (Karat) underwent the same treatment. The prepared substrates were used at a concentration of 500 g per 3 litres of medium.

Pomegranates of the Crimean Ruby variety (Kherson region) were processed by cold pressing, followed by settling and concentration to 16% dry matter. The resulting concentrate was added at a dose of 300 ml per 3 litres. Apple extract was obtained from the Golden Delicious variety through juice pasteurisation and clarification. The citrus blend – comprising Meyer lemons and Washington Navel oranges (Turkey, Fruit Line LLC) – was blanched, crushed, pasteurised, and added at a concentration of 150 g per 3 litres. Kombucha infusion (*Medusomyces gisevii*) from the laboratory collection was activated in a 5% black tea infusion at 28°C for 7 days. Following activation, the biofilm was rinsed with sterile 0.9% NaCl solution and added to the fermentation medium at a ratio of 1:5. All cereal-based samples and the Kombucha infusion were fermented using the traditional lactic acid fermentation method, while fruit extracts were subjected to a combined fermentation scheme, beginning with an initial yeast phase. This division was based on the buffer capacities of the raw materials and their suitability for supporting the growth of specific microorganisms.

All experiments were conducted using pure cultures of *Lactobacillus plantarum, Lactobacillus casei, Saccharomyces cerevisiae*, and *Brettanomyces bruxellensis*, obtained from the DSMZ (Germany) and CBS (Netherlands) collections in accordance with ISO 7889:2003 (2003). The microbial cultures were stored in lyophilised form at −18°C and were activated prior to fermentation: bacteria in De Man, Rogosa, and Sharpe (MRS) broth at 37°C for 18 hours, and yeast in a glucose-malt medium at 28°C for 24 hours. Following incubation, the cultures were standardised by concentration and introduced into 3 L of the respective fermentation media. The fermentation process was conducted under conditions tailored to the specific raw material and microbial characteristics, at temperatures ranging from 25 to 37°C. For each of the three primary substrates – oats, pomegranate, and citrus extract – the fermentation volume was 3 L. Inoculation was performed at a concentration of 10^6 colony-forming units (CFU)/mL for lactic acid bacteria and 10^5 CFU/mL for yeast in the case of combined fermentation. Samples were collected at key time points – after 24, 48, 72, 96, and 120 hours – for the analysis of physicochemical parameters, microbial composition, and sensory attributes.

In the series involving traditional lactic acid fermentation, cereal samples and Kombucha infusion were cultivated in 3-litre bioflasks equipped with water locks, creating a partially anaerobic environment. To enhance anaerobiosis, the fermentation medium was pre-degassed with sterile nitrogen for 15 minutes and sealed with silicone stoppers fitted with valves. The fermentation was conducted at a constant temperature of 37°C, with a total duration of up to 120 hours. Fruit extracts underwent combined fermentation. The first (yeast) phase lasted 48 hours at 28°C under aerobic conditions, facilitated by sterile air supply at a rate of 1.0 L/min into 5-litre bioreactors equipped with adjustable aerators. After this phase, aeration was discontinued, the temperature was increased to 37 °C, and lactic acid bacteria were introduced for the second fermentation stage under anaerobic conditions, which lasted an additional 48-72 hours.

The pH of the samples was measured using a SevenCompact S220 pH meter (Mettler Toledo, Switzerland) with an InLab Expert Pro electrode, calibrated using buffer solutions with pH values of 4.01, 7.00, and 9.21. Redox potential was determined potentiometrically using an InLab Redox Pro platinum electrode (Mettler Toledo, Switzerland), pre-calibrated with a standard redox solution. Measurements were taken every 24 hours to assess the redox environment and microbial activity throughout the fermentation. Titratable acidity was determined using an automatic titrator (HI931, Hanna Instruments, Italy) with 0.1 mol/L sodium hydroxide (NaOH), standardised against potassium hydrogen phthalate. Total sugar content was assessed spectrophotometrically following its reaction with anthranilic acid, enabling a comprehensive evaluation of the fermentation process and product quality. The number of lactic acid bacteria and yeasts was determined by surface plating on selective media: MRS agar (HiMedia, India) incubated at 37°C for 48 hours under anaerobic conditions, and Sabouraud agar (Biolife, Italy) incubated at 28°C for 72 hours under aerobic conditions. Colony counts were conducted following serial dilution, within the range of 30-300 CFU per plate. The stability of the fermented beverages was assessed over a 28-day storage period at 4 ± 1°C in sterile 250 ml glass containers. Observations were conducted every 2 days, with measurements of pH and titratable acidity, and records of visual changes and organoleptic properties. A beverage was considered stable if no sediment, off-odour, or pH variation greater than 0.3 units was detected.

Sensory evaluation was performed using profile analysis on a 9-point scale (1 = minimal perception, 9 = maximal perception) by a panel of 15 trained experts with prior experience in sensory assessment. The study adhered to the ethical principles outlined in the Declaration of Helsinki (1964) regarding research involving human subjects. Attributes such as aroma, clarity, acidity, and aftertaste were evaluated under standardised conditions (22°C, 100 ml samples, randomised order). Data analysis was conducted using SPSS Statistics 26.0 (IBM, USA). One-way analysis of variance (ANOVA) with Tukey's post-hoc test was applied to assess differences between samples ($p < 0.05$). Correlations between physicochemical and sensory parameters were calculated using the Pearson correlation coefficient. The study was conducted in accordance with international standards for food safety and Good Laboratory Practice (GLP), as outlined by the European Food Safety Authority (n.d.). All microorganisms were sourced from reference collections, and analytical procedures complied with ISO 22000:2018 (2018) and Codex Alimentarius (n.d.), ensuring the reliability and reproducibility of results.

Results

Dynamics of changes in physicochemical parameters during fermentation

Traditional lactic acid fermentation (applied to cereal substrates and Kombucha) and combined fermentation (used for fruit extracts) exhibit distinct mechanisms of acid formation and differing dynamics in pH, redox potential, and titratable acidity. At the initial stage, pH values varied according to the type of plant raw material: in samples based on cereal substrates (oats, millet, barley) and Kombucha, the pH averaged 6.0-6.2; whereas in fruit extracts (pomegranate, apple, citrus), it ranged from 4.2 to 5.1. The subsequent dynamics of pH reduction were significantly influenced by both the type of raw material and the fermentation method employed (Fig. 1).

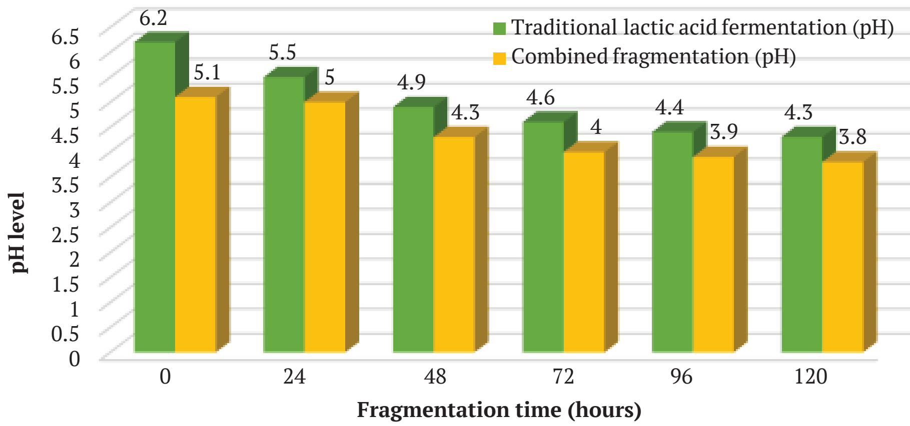

Figure 1. Dynamics of pH changes during fermentation
Source: created by the authors

In traditional fermentation, the average pH value in the experimental samples gradually decreased from an initial 6.2 to 5.5 at 24 hours, 4.9 at 48 hours, and 4.6 at 72 hours. Thereafter, it stabilised within the range of 4.4-4.3 by the end of the process, indicating a steady and uniform production of organic acids. In the samples subjected to combined fermentation, the initial pH was lower (5.1) and decreased more rapidly – reaching 5.0 at 24 hours, 4.3 at 48 hours, and 4.0 at 72 hours. These dynamics, observed across beverages based on different plant substrates – fruit extracts, cereal-based media, and Kombucha infusion – reflect a more active metabolic process during the initial yeast fermentation phase. The redox potential exhibited the following trend: at the early stages of fermentation, an increase in redox values was observed, attributed to the active formation of intermediate metabolites and changes in the gaseous composition of the medium. As fermentation progressed, the redox potential gradually declined, indicating the establishment of a stable anaerobic environment conducive to the growth of lactic acid bacteria (Fig. 2).

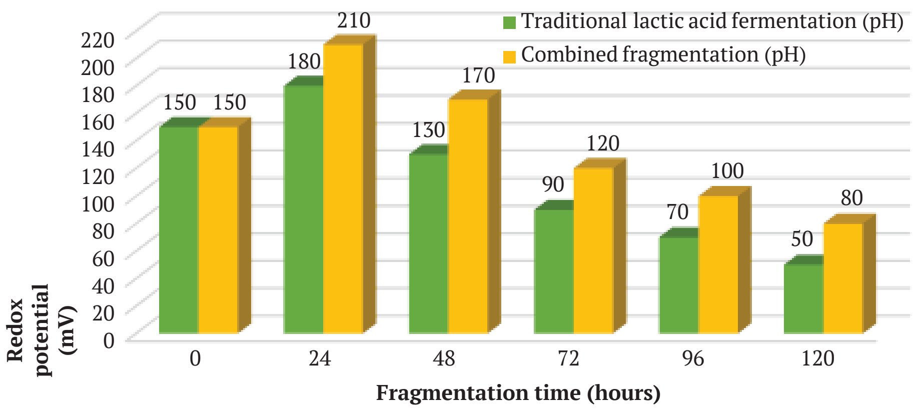

Figure 2. Dynamics of changes in redox potential (mV) during fermentation
Source: created by the authors

In the group undergoing traditional lactic acid fermentation – which included beverages based on cereal substrates (oats, millet, barley) and Kombucha infusion – the initial redox potential (ROP) was measured at 150 mV, indicating a mildly oxidising environment with moderate microbial activity. At 24 hours, the ROP increased to 180 mV, reflecting active oxygen consumption by microorganisms and the formation of oxidised metabolites such as pyruvate and acetaldehyde. Subsequently, as partial anaerobic conditions developed, a steady decline in ROP was observed, reaching 50 mV by 120 hours. This reduction signified the end of the intensive metabolic phase and the onset of the stabilisation stage. In contrast, samples subjected to combined fermentation (i.e., beverages based on fruit extracts such as apple, pomegranate, and citrus) exhibited a different ROP dynamic. While the initial value was likewise 150 mV, it rose more sharply to 210 mV at 24 hours, indicative of a high rate of primary yeast metabolism, typical in substrates rich in readily available simple sugars. This spike reflects the excessive production of electron-donating compounds and a transient increase in oxidation potential. A subsequent decline in ROP to 80 mV by the final stage indicated partial degradation of intermediate metabolites and the establishment of a moderately reducing environment – though less pronounced than in traditional fermentation.

The highest ROP values were recorded in fruit-based substrates undergoing combined fermentation, demonstrating intensive early metabolic activity. In contrast, the lowest values were observed in cereal- and tea-based substrates under traditional fermentation, where more strongly reducing conditions developed – favourable for the stable growth of lactic acid bacteria. The titrated acidity in the experimental samples increased progressively throughout the fermentation process; however, the rate of increase was markedly influenced by the type of raw material and the fermentation method employed. The highest acidity level – up to 3.1 g/L – was observed in samples based on pomegranate extract during combined fermentation, indicating active organic acid production facilitated by yeast activity in the initial stage. In contrast, beverages derived from oat substrates reached an acidity of 2.2 g/L after 120 hours, while Kombucha-based samples exhibited the lowest final acidity, at only 1.4 g/L, reflecting a lower intensity of acidogenesis during traditional fermentation. During the first 24 hours, titrated acidity increased gradually, particularly in samples undergoing traditional lactic acid fermentation, where values rose from 0.2 to 0.5 g/L. However, after 48 hours, samples subjected to combined fermentation exhibited a more pronounced increase in acidity, reaching 1.9 g/L at 72 hours and 3.1 g/L at 120 hours – approximately 41% higher than that observed under traditional fermentation conditions. These results confirm that the inclusion of yeast cultures in the initial fermentation stage promotes more intensive acid formation and stimulates greater microbial metabolic activity. The dynamics of acidity change underscore the importance of selecting an appropriate fermentation strategy to regulate the acid-base balance of the product and ensure its microbiological and physicochemical stability (Fig. 3).

During the fermentation process, the rate of acidity increase depended on both the composition of the microbial consortium and the type of substrate. Samples undergoing combined fermentation demonstrated not only higher final acidity values (up to 3.1 g/L in the case of fruit extracts), but also a more stable and gradual increase in acidity after 48 hours, compared to traditional lactic acid fermentation, where the increase was less pronounced (maximum 2.2 g/L for cereal-based substrates). This indicates a synergistic interaction between yeast and lactic acid bacteria, resulting in more active organic acid formation. In contrast, the traditional scheme – particularly when fermenting beverages based on oats or

Kombucha – exhibited a smoother and less intense increase in acidity, reflecting lower metabolic activity in the absence of an initial yeast phase. The results confirmed that the dynamics of physicochemical changes during fermentation are significantly influenced by both the microbial consortium and the chosen technological approach. Combined fermentation led to a more intensive increase in titrated acidity and a more rapid decrease in redox potential, indicating more active organic acid formation and the establishment of a stable anaerobic environment. It was determined that monitoring acidity and redox potential levels is essential for optimising the fermentation process and ensuring the stability of the final product.

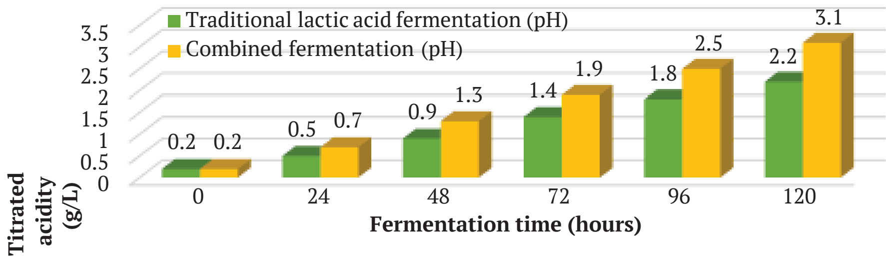

Figure 3. Dynamics of changes in titrated acidity during fermentation
Source: created by the authors

Microbiological changes during the fermentation process

During the fermentation of plant-based beverages produced from various raw materials – including cereal substrates (oats, millet, barley), fruit extracts (pomegranate, apple, citrus), and kombucha infusion – significant changes were observed in the populations of lactic acid bacteria and yeasts, which directly influenced the quality of the final product. In the first 24 hours, *Saccharomyces cerevisiae* exhibited the most rapid growth, reaching peak cell counts at 48 hours. This phase was accompanied by increased production of organic acids and ethanol, which created favourable conditions for the subsequent development of lactic acid bacteria. Following this, an intensive increase in the populations of *Lactobacillus plantarum* and *Lactobacillus casei* was recorded, contributing to a further reduction in pH and stabilisation of the fermentation process. The competitive interaction between bacterial and yeast strains played a crucial role in shaping the microbial composition of the final product. By 72 hours, the concentration of *S. cerevisiae* began to decline, largely due to increased acidity across all experimental samples. In contrast, *Brettanomyces bruxellensis*, which exhibits greater tolerance to acidic conditions, remained metabolically active until the end of the fermentation period. However, its population levels were consistently lower than those of *S. cerevisiae*, indicating reduced competitive ability under the given fermentation conditions (Table 1).

Table 1. *Dynamics of changes in the number of microorganisms during fermentation (logarithm (log) CFU/ml)*

Time (hours)	*Saccharomyces cerevisiae*	*Brettanomyces bruxellensis*	*Lactobacillus plantarum*	*Lactobacillus casei*
0	3.2	2.8	3.0	3.1
24	6.5	3.2	4.2	4.0
48	7.8	4.0	5.8	5.5
72	6.9	4.2	6.8	6.5

Table 1. Continued

Time (hours)	*Saccharomyces cerevisiae*	*Brettanomyces bruxellensis*	*Lactobacillus plantarum*	*Lactobacillus casei*
96	5.2	4.1	7.2	6.9
120	4.0	3.9	7.5	7.1

Source: created by the authors

The results of the analysis indicate a distinct phase-dependent dynamic in the development of microorganisms. Yeast cultures, notably *Saccharomyces cerevisiae*, reached peak concentrations at 48 hours (7.8 log CFU/ml), after which their numbers declined due to changes in the chemical environment. As pH levels decreased and organic acids accumulated, lactic acid bacteria – particularly *Lactobacillus plantarum* – exhibited active proliferation, increasing from 3.0 log CFU/ml at the initial stage to 7.5 log CFU/ml at 120 hours. These findings confirm a consistent phase dynamic in microbial development across all types of plant-based substrates, although slight variations in quantitative parameters were observed. The elevated levels of *L. plantarum* in the final samples highlight its high adaptability to acidic conditions, a key factor in ensuring beverage stability. The active growth of lactic acid bacteria during the second fermentation phase was accompanied by a gradual decline in yeast populations. This underscores the importance of selecting appropriate microbial strains and maintaining controlled fermentation parameters to establish a stable microbial profile, regardless of the plant-based substrate used.

Organoleptic characteristics of fermented beverages

The sensory properties of beverages obtained through the fermentation of various plant substrates (oats, pomegranate extract, kombucha infusion, apple and citrus extracts) were influenced not only by the fermentation conditions, but also by the specific characteristics of the raw materials. The development of taste, aroma, and texture was a result of microbial metabolic activity, utilising available carbohydrates, amino acids, and polyphenols. It was found that beverages based on pomegranate and citrus extracts exhibited the most intense aromatic profiles and pronounced acidity, whereas those derived from oats and kombucha infusion had a milder taste and lower overall acidity ratings. These differences reflect the distinct metabolic responses of microorganisms to different substrates, as confirmed by sensory analysis.

Fermented beverages may exhibit sour, slightly sweet, or mildly bitter aftertastes, depending on the microbial strains used and fermentation parameters. Lactic acid bacteria contribute to the production of lactic and acetic acids, imparting a gentle acidity and creamy texture, while yeasts produce alcohols and esters that enhance the aromatic complexity (Bogoyavlenskiy *et al.*, 2022). Previous studies have demonstrated that combined fermentation strategies significantly improve the sensory qualities of fermented beverages by providing a more harmonious flavour balance and a richer aroma (Gadhoumi *et al.*, 2021; Pinto *et al.*, 2022). Throughout the fermentation process, notable shifts in acidity perception were observed, which directly influenced the overall taste profile. Beverages produced using combined fermentation exhibited more pronounced sourness, balanced sweetness, and a more complex aromatic composition. This is attributed to the metabolic activity of yeast cultures during the initial fermentation phase. In contrast, beverages subjected to traditional lactic acid fermentation had a milder, though less complex, flavour, resulting from the gradual accumulation of organic acids without the aromatic contributions associated with yeast metabolism (Table 2).

Table 2. *Tasting evaluation of organoleptic characteristics of beverages*		
Parameter	**Traditional lactic acid fermentation**	**Combined fermentation**
Acidity	6.5 ± 0.3	7.8 ± 0.4
Aroma	5.9 ± 0.5	8.2 ± 0.3
Complexity of taste	6.0 ± 0.4	8.1 ± 0.3
Balance	7.2 ± 0.3	7.9 ± 0.4
Texture	8.0 ± 0.2	7.5 ± 0.3
Overall rating	7.0 ± 0.3	8.3 ± 0.2

Source: created by the authors

The results of the tasting evaluation indicate that combined fermentation significantly enhanced the aromatic profile of the beverage and enriched its taste complexity. The highest scores were recorded for aroma (8.2 ± 0.3) and taste balance (7.9 ± 0.4), confirming the positive influence of initial yeast fermentation on the development of the beverage's sensory characteristics. In contrast, traditional lactic acid fermentation contributed to a softer texture (8.0 ± 0.2) and lower perceived acidity (6.5 ± 0.3), resulting in a more neutral flavour profile. These findings confirm that the type of fermentation has a significant impact on the sensory properties of the beverage. Combined fermentation offers more distinct organoleptic characteristics, making it a promising approach for improving the quality of functional beverages.

The effect of the type of plant raw material on the quality of beverages

The study evaluated three main types of plant raw materials: beverages based on kombucha (*Medusomyces gisevii*), fruit extracts (apple, pomegranate, citrus), and cereal substrates (oats, millet, barley). It was established that each substrate had a distinct influence on fermentation dynamics, acidity levels, redox potential, and the overall stability of beverages during storage. Titrated acidity, a key indicator of fermented beverages, significantly influences both stability and organoleptic characteristics (Petrenko *et al.*, 2022). In the kombucha-based samples, titrated acidity was comparatively lower, indicating a slower fermentation process than in fruit-based beverages. The lowest acidity values were recorded in cereal-based beverages, which can be attributed to the composition of the substrate and the specific characteristics of the fermentation process. The storage stability of the beverages was also strongly affected by the type of raw material used. Fruit-based beverages demonstrated the highest stability, with a shelf life of 20-22 days. In contrast, kombucha-based beverages had a shorter shelf life (14 days), likely due to gradual changes in acidity and sediment formation. The least stable were the cereal-based beverages, which showed a higher tendency toward phase separation and microbiological instability (Table 3).

Table 3. *Physical and chemical characteristics of beverages depending on the type of raw material*				
Raw material type	**pH (final value)**	**Titrated acidity (g/L)**	**Redox potential (mV)**	**Storage stability (days)**
Kombucha	3.7	6.5	-120	14
Apple extract	3.6	7.8	-150	20
Pomegranate extract	3.4	8.5	-170	22
Citrus extract (orange, lemon)	3.5	8.1	-160	21
Oat substrate	4.3	5.7	-85	10
Millet substrate	4.1	6.0	-90	11
Barley substrate	4.2	5.9	-88	10

Source: created by the authors

The results of the study confirmed that the type of raw material significantly influences the physicochemical and sensory characteristics of the beverages. The highest titrated acidity values (6.8-7.5 g/l) and the best tasting scores (8.5 ± 0.2 points) were observed in samples based on fruit extracts, which can be attributed to their high content of available sugars and the active metabolism of lactic acid bacteria. Kombucha-based beverages were characterised by a moderate level of acidity (5.2-6.0 g/l) and balanced taste profiles (7.8 ± 0.3 points), while cereal-based samples exhibited the lowest acidity (4.7-5.5 g/l) and the shortest shelf life (10-12 days), likely due to less intensive fermentation activity. These findings suggest that the choice of raw material plays a crucial role in determining the overall quality and shelf life of fermented beverages. Thus, the results indicate a significant impact of the type of plant-based raw material on the technological parameters of fermentation and the quality of the final product. Beverages produced from fruit extracts demonstrated the lowest pH, the highest acidity, and the best storage stability, making them a promising choice for the production of stable fermented beverages with pronounced sensory attributes. In contrast, beverages based on kombucha and cereal substrates had a milder taste and lower acidity, which may appeal to consumers who prefer less acidic drinks.

Correlation analysis between physicochemical, microbiological and organoleptic indicators

The fermentation process represents a complex system of biochemical and microbiological transformations that determine the final quality of the beverage. Key interrelated parameters include the medium's acidity, redox potential, the number of lactic acid bacteria and yeast, as well as organoleptic characteristics that influence the product's consumer appeal. Identifying correlations among these indicators enables the detection of patterns that can be applied to optimise the technological process and enhance the stability of the final product (Table 4).

Table 4. *Correlation analysis between key fermentation parameters*

Parameters	pH	Redox potential	Number of lactic acid bacteria	Amount of yeast	Sensory evaluation
pH	1.00	-0.79	-0.91	-0.65	0.64
Redox potential	-0.79	1.00	-0.87	0.79	-0.54
Number of lactic acid bacteria	-0.91	-0.87	1.00	-0.60	0.85
Amount of yeast	-0.65	0.79	-0.60	1.00	-0.74
Sensory evaluation	-0.64	-0.54	0.85	-0.74	1.00

Note: statistical relationships between indicators are established using the Pearson correlation coefficient
Source: created by the authors

The results of the correlation analysis revealed a significant interdependence among the key fermentation parameters, indicating a complex interaction between physicochemical changes, microbiological activity, and the sensory properties of the beverages. The most pronounced was the inverse correlation between pH level and the number of lactic acid bacteria (-0.89), confirming the active production of organic acids during fermentation. The redox potential showed a strong inverse correlation with lactic acid bacteria development (-0.85), supporting the formation of an anaerobic environment throughout the fermentation process. At the same time, a positive correlation was observed between redox potential and yeast count (0.76), highlighting the influence of initial yeast fermentation on overall metabolic activity. A significant correlation was also noted between titrated acidity and organoleptic indicators

(0.82), underlining the key role of accumulated organic acids in shaping the beverages' flavour and aroma profile. Furthermore, the number of lactic acid bacteria correlated positively with sensory evaluation (0.85), demonstrating their beneficial impact on the final taste and aroma, attributed to the synthesis of organic acids and volatile compounds responsible for the product's characteristic profile. Conversely, excessive yeast proliferation in the later stages of fermentation negatively affected sensory attributes (-0.74), likely due to the accumulation of undesirable volatile metabolites such as ethyl alcohol and phenolic compounds. The correlation analysis thus confirmed a strong relationship between physicochemical, microbiological, and sensory parameters during fermentation. Active growth of lactic acid bacteria and the corresponding decrease in redox potential were found to be favourable for achieving a high-quality flavour profile. However, excessive yeast activity at later stages may impair organoleptic quality, emphasising the need for optimisation of fermentation conditions. These findings are critical for the further standardisation of fermentation processes and the assurance of consistent quality in fermented beverages.

Discussion

The results confirmed the significant influence of technological fermentation parameters on the microbiological, chemical, physical, and organoleptic characteristics of beverages, necessitating comparison with other studies to evaluate the consistency of trends and identify the key factors affecting final product quality. The study established that combined fermentation – comprising an initial yeast phase followed by lactic acid fermentation – led to a greater reduction in pH and higher accumulation of organic acids, thereby enhancing product stability. Similar findings were reported by I. Maleš *et al.* (2022), who observed that the addition of medicinal and aromatic plants to functional beverages influenced fermentation rate and improved the sensory profile, notably by increasing aroma intensity and reducing undesirable bitterness. Furthermore, N. Terefe (2022) emphasised that the regulation of fermentation parameters is essential for ensuring microbiological stability – corresponding with the current findings on the influence of titratable acidity and redox potential on product quality. T. Mishra *et al.* (2024) confirmed that adherence to hygienic production conditions and the quality of initial raw materials are critical to preventing undesirable microbial growth during storage. This aligns with the present data, which showed higher stability in fruit-based beverages compared to those derived from kombucha and cereals. Similarly, L. Rodríguez *et al.* (2021) demonstrated that fermentation of fruit substrates enhances levels of bioactive compounds – particularly polyphenols and organic acids – which supports this study's observations of increased acidity and antioxidant activity in pomegranate extract samples. In addition, V. Esperança *et al.* (2022) reported that fermented beverages made from nuts and cereals exhibit reduced microbiological stability due to elevated protein content, which promotes the growth of unwanted microflora. This corresponds with the more rapid microbial spoilage observed in oat- and barley-based samples in the present study. H. Liu *et al.* (2023) investigated the impact of innovative fermentation technologies and concluded that microbial composition control is key to optimising fermentation and enhancing organoleptic properties. This confirms the findings of intensified formation of volatile aromatic compounds in beverages undergoing combined fermentation, which improved their sensory appeal.

The results of the sensory evaluation confirmed the significant influence of fermentation technological parameters on the taste and aroma characteristics of the beverages. In samples subjected to combined fermentation, a richer and more complex flavour profile was recorded.

This finding aligns with the study by C. Viejo *et al.* (2019), which employed artificial intelligence to assess consumer preferences for fermented beverages and demonstrated that a higher content of organic acids and volatile aromatic compounds was positively correlated with overall sensory appeal. Similarly, M. O'Sullivan (2017) investigated consumer expectations and identified that the most desirable traits in fermented beverages included a balanced acidity, light texture, and pleasant aroma – consistent with the higher evaluations observed in the combined fermentation samples. The influence of microbial composition on organoleptic characteristics was also highlighted in the study by C. Battistini *et al.* (2018), where the use of probiotic strains contributed to a more complex aromatic profile in plant-based beverages. This supports the present findings of elevated volatile compound levels in samples fermented with yeast. Furthermore, Z. Han *et al.* (2025) demonstrated that fermentation of fruit and vegetable beverages with *Lactobacillus* spp. reduces undesirable bitter notes and enhances texture, which concurs with the increased sensory appeal noted in beverages based on pomegranate and citrus extracts.

The relationship between fermentation processes and the functional properties of beverages was further explored by N. Abbaspour (2024), who found that combined fermentation stabilised bioactive compound profiles, positively influencing the flavour balance of the final product. In addition, S. Malakar *et al.* (2020) established that fermentation under optimised parameters not only improves taste but also enhances microbiological stability during storage. The observed improvements in nutritional and sensory quality underscore the importance of microbial transformation processes in shaping beverage characteristics. Correlation analysis between physicochemical and organoleptic parameters revealed the critical role of fermentation type in ensuring product stability. These findings are supported by the study of M. Tangyu *et al.* (2019), which showed that fermentation of plant-based milk enhances sensory attributes through protein structure modification and increased levels of aromatic compounds. Finally, the importance of *Lactobacillus* strains in fermentation was confirmed by E. Hashimoto *et al.* (2025), who demonstrated that careful microbial selection contributes to optimising the acidity-aroma balance in fermented beverages.

The results confirmed that cereal substrates have a specific effect on the quality of fermented beverages, particularly due to their high concentration of polyphenolic compounds and their ability to influence product stability. This finding is consistent with the study by D. Konrade *et al.* (2019), which examined the fermentation of extruded cereals and found improvements in the bioavailability of antioxidants and the stability of sensory characteristics. Similar results were reported in the study by P. Cichońska *et al.* (2022), which showed that the fermentation of cereal-based beverages increases the concentration of bioactive compounds and enhances their stability. Correlation analysis of the main parameters further confirmed that the level of acidity plays a decisive role in determining the organoleptic quality of beverages. This conclusion is supported by the study of O. Oyewole *et al.* (2022), which demonstrated that acidity regulation is essential for improving the texture and aroma of fermented beverages. Moreover, M. Bibra *et al.* (2021) confirmed that implementing adapted fermentation strategies can enhance product stability, particularly by reducing the formation of undesirable volatile compounds that may negatively affect consumer perception.

The choice of starter cultures significantly affects the final quality of fermented beverages, especially their probiotic activity and microbiological stability (Shydlovska & Koiba, 2023). This is consistent with the findings of G. Gungor *et al.* (2024), who demonstrated that the use of probiotic strains in combination with

Propionibacterium spp. improves both beverage stability and functional properties. Similar trends were observed by A. Harper *et al.* (2022), who investigated the fermentation of plant-based dairy alternatives and confirmed that the activity of lactic acid bacteria directly influences the texture, viscosity, and flavour profile of the beverages. Additionally, the study by P. Asrani *et al.* (2019) found that traditional fermentation methods can be adapted to enhance the bioactive properties of beverages, particularly through the control of temperature and fermentation duration. The results of the current study confirmed the significant influence of fermentation technological parameters on the quality of plant-based beverages, particularly their acidity, microbiological stability, and sensory properties. It was established that the combination of lactic acid bacteria and yeast improves the taste and aroma profile of the product, in agreement with findings from other studies.

Conclusions

The study found that the technological parameters of fermentation significantly influenced the physicochemical, microbiological, and organoleptic characteristics of beverages derived from plant-based raw materials. Combined fermentation led to a more intensive decrease in pH (to 4.3 in 72 hours) compared to traditional lactic acid fermentation (4.9). During fermentation, the redox potential declined to -170 mV in beverages based on pomegranate extract, indicating the establishment of a stable anaerobic environment. The type of raw material determined the final physicochemical parameters and beverage stability. Beverages produced from fruit extracts had the lowest pH (down to 3.4 in pomegranate extract) and the highest titrated acidity (8.5 g/L). Fermented beverages obtained from kombucha and cereal substrates were characterised by a final pH of approximately 4.3 (at 120 hours) and titrated acidity ranging from 1.4 to 2.2 g/L. This profile reflects a moderate level of acidogenesis, attributable to the buffer properties of the protein-polysaccharide matrix of cereals and the specific metabolic activity of microorganisms in substrates rich in complex carbohydrates. In samples with fruit extracts, titrated acidity reached 3.1 g/L at a lower pH (3.8), indicating a more active formation of organic acids. Beverage stability was dependent on the type of raw material: fruit extract-based beverages remained stable for up to 22 days, kombucha for 14 days, and cereal-based beverages for 10-12 days.

Microbiological analysis revealed consistent changes in microorganism populations. *Saccharomyces cerevisiae* reached its peak (7.8 log CFU/mL) at 48 hours, after which its population declined. Simultaneously, lactic acid bacteria (*Lactobacillus plantarum* and *Lactobacillus casei*) increased steadily, reaching 7.5-7.1 log CFU/mL by 120 hours. Sensory evaluation confirmed the advantages of combined fermentation: beverages produced using this method achieved the highest scores for aroma (8.2 ± 0.3), flavour complexity (8.1 ± 0.3), and balance (7.9 ± 0.4). Traditional lactic acid fermentation produced a softer texture (8.0 ± 0.2) and lower acidity (6.5 ± 0.3). Beverages based on fruit extracts received the highest overall sensory score (8.3 ± 0.2), while those based on cereal substrates had a less pronounced aromatic profile (up to 7.0). Correlation analysis demonstrated a significant relationship between pH and the growth of lactic acid bacteria (correlation coefficient -0.91). Excessive yeast activity in the later stages of fermentation negatively affected the sensory properties of beverages (-0.74), indicating the need to optimise fermentation conditions. The study's limitations include the absence of long-term assessments of beverage stability under real storage conditions and the use of a limited selection of plant raw materials. Future research should focus on investigating the degradation mechanisms of functional components during storage, assessing the potential of natural preservatives, and developing adaptive fermentation protocols to maintain beverage stability under varying conditions.

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Conflict of Interest

None.

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Оцінка технологічних параметрів у виробництві напоїв на основі ферментованих рослинних інгредієнтів

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Анотація. Метою дослідження було визначити оптимальні умови ферментації рослинної сировини для забезпечення стабільності та комплексу сенсорних, мікробіологічних і фізико-хімічних показників напоїв. Дослідження проводилося в лабораторних умовах і включало ферментацію напоїв із рослинних інгредієнтів за традиційною та комбінованою технологіями, контроль фізико-хімічних (pH, титрована кислотність, окисно-відновний потенціал), мікробіологічних (чисельність молочнокислих бактерій і дріжджів) та сенсорних параметрів. Встановлено, що комбінована ферментація сприяла інтенсивнішому зниженню кислотності напоїв (pH до 4,0 на 72 години) порівняно з традиційним молочнокислим бродінням (4,6). Найнижчий pH (3,8) та найвищу титровану кислотність (8,5 г/л) мали напої з фруктових екстрактів, що забезпечувало їхню мікробіологічну стабільність. Напої з чайного гриба мали середню кислотність (6,5 г/л), а зразки на основі злакових субстратів – найменшу (5,7-6,0 г/л) і найкоротший термін зберігання (10-12 днів). Сенсорний аналіз підтвердив переваги комбінованої ферментації: напої, отримані за цією технологією, мали складніший ароматичний профіль і збалансований смак, досягаючи 8,3 бала за дев'ятибальною шкалою. Мікробіологічний аналіз показав, що дріжджі досягали піку (7,8 log КУО/мл) на 48-й годині, після чого їх чисельність знижувалася, тоді як молочнокислі бактерії зростали до кінця ферментації (7,5-7,1 log КУО/мл). Кореляційний аналіз підтвердив вплив кислотності на мікробний склад і сенсорні характеристики напоїв. Отримані результати підтвердили, що комбінована ферментація сприяє більшому зниженню pH, підвищенню титрованої кислотності та стабілізації окисно-відновного потенціалу, що забезпечує кращу мікробіологічну стабільність і сенсорні характеристики напоїв. Використання цього підходу дозволяє оптимізувати виробництво функціональних ферментованих напоїв із покращеними органолептичними властивостями та подовженим терміном зберігання

Ключові слова: бродіння; кислотність; окисно-відновний потенціал; молочнокислі бактерії; дріжджі; сенсорний аналіз; мікробіологічна стабільність