EXPERIMENTAL ARTICLES

UDC 579.6:579.864.1 https://doi.org/10.15407/biotech19.01.027

PROBIOTIC PROPERTIES OF LACTIC ACID BACTERIA ISOLATED FROM SOUR BREWING WORT

O.S. Putria[1]
A.I. Romanenko[1]
S.A. Marushchenko[1]
P.R. Zubyk[1]
A.D. Khablenko[1,2]
S.G. Danylenko[2]

[1]National Technical University of Ukraine "Igor Sikorsky Kyiv Polytechnic Institute"

[2]Institute of Food Resources of the National Academy of Agrarian Sciences of Ukraine

E-mail: khablenkoad@gmail.com

The study of the probiotic potential of lactic acid bacteria isolated from sour brewing wort is a promising direction for developing new functional fermented products, as these strains may exhibit enhanced antagonistic activity and improved survival under stress conditions.

The work aims to isolate, identify, and determine selected probiotic properties of lactic acid bacteria derived from sour brewing wort.

Methods. The following method was used to prepare the brewing wort. Isolation of lactic acid bacteria (LAB) strains was carried out from brewing wort that had been spontaneously fermented for 12 months. Identification was performed using physiological and biochemical analyses of LAB metabolic characteristics. Antagonistic activity of the isolate was assessed using the well-diffusion method, antibiotic susceptibility by the disk-diffusion method, and adhesive properties by spectrophotometry. Enzymatic activities were determined qualitatively.

Results. A culture of *Lactiplantibacillus pentosus* isolated from brewing wort was identified. Its strong antagonistic effect against the test culture, *S. aureus*, was demonstrated, with an inhibition zone of 30.0 ± 0.10 mm. Resistance to aminoglycosides and penicillins was established, consistent with the typical antibiotic susceptibility profile of LAB. Adhesion indicators — autoaggregation and hydrophobicity — were 44.80 ± 2.62% and 46.20 ± 3.00%, respectively. No studied enzymatic activities were observed.

Conclusions. *Lpb. pentosus* isolated from sour brewing wort exhibits antagonism toward *S. aureus*, resistance to aminoglycosides, penicillins, and glycopeptides, moderate autoaggregation, and high cell wall hydrophobicity. The studied enzymatic activities were not detected. The results indicate potential probiotic activity and the need for further studies on stress tolerance.

Keywords: lactic acid bacteria, sour wort, *Lactiplantibacillus pentosus*, antagonism, adhesive properties, enzymatic activity, probiotics, biotechnology.

Lactic acid bacteria (LAB) are among the most extensively studied and most numerous groups of microorganisms [1]. They are known for their antagonism toward a broad range of pathogenic organisms, including antifungal effects [2], as well as immunomodulatory, anticancer, antioxidant, anti-inflammatory, and other activities [3]. This beneficial impact on the host organism makes them promising candidates for consideration as probiotic organisms [4].

Despite this, the primary role of LAB lies in their ability to ferment various

Citation: Putria, O. S., Romanenko, A. I., Marushchenko, S. A., Zubyk, P. R., Khablenko, A. D., Danylenko, S. G. (2026). Probiotic properties of lactic acid bacteria isolated from sour brewing wort. *Biotechnologia Acta*, 19(1), 27–37. https://doi.org/10.15407/biotech19.01.027

macromolecules, which is determined by their well-developed metabolic pathways and metabolic flexibility [5]. This characteristic encourages the consideration of different food products as sources for LAB isolation. Typical sources include spontaneously fermented dairy products [6, 7]. Publications have also reported the isolation of LAB from spontaneously fermented sausages (*Lactiplantibacillus* spp., *Pediococcus* spp.) [8–10] and seafood (*Leuconostoc* spp., *Carnobacterium* spp.) [11]. Considerable attention is likewise given to plant raw materials, particularly food-industry by-products such as pomace, pulp, press cakes, and others [12]. For example, *Lactiplantibacillus plantarum* was isolated from rose hip pomace [13], while silage yielded *Leuconostoc* spp., *Lactococcus* spp., *Enterococcus* spp., *Pediococcus* spp., *Lactobacillus* spp., and others [14, 15]. Sugarcane [16], millet [17], and other herbaceous plants [18] have also been used for LAB isolation.

Beverages of various origins are considered promising sources for isolating LAB strains with probiotic properties. Traditional local low-alcohol fermented drinks are most commonly used for this purpose, such as *boza*, *pozol*, *togwa*, and others [7, 18]. From the Ethiopian cereal-fermented drink *tella*, *Pediococcus pentosaceus*, *Lactobacillus curvatus*, *Leuconostoc mesenteroides*, and *Lpb. plantarum* were isolated and identified [19]. The use of the traditional African beer *Chibuku* enabled the recovery of representatives of the genera *Lactobacillus*, *Lactococcus*, *Leuconostoc*, *Streptococcus*, and *Enterococcus* [20]. Despite this, only a small number of publications address LAB isolation specifically from intermediate brewing products (mash or sour wort) [21]. Therefore, the present study aimed to isolate and identify lactic acid bacteria from sour beer wort and to assess their probiotic properties.

Materials and Methods

Isolation of lactic acid bacteria (LAB) strains was carried out from sour beer wort. Barley malt *Premium Pilsner* (Weyermann) was used to prepare the wort. The wort was prepared using the infusion mashing method, with a stepwise increase in temperature and incubation at the following temperature rests: acid rest — 35 °C for 30 min; protein rest — 45 °C for 45 min; maltose rest — 65 °C for 45 min; mash-out — 78 °C for 15 min [22]. The sugar content of the cooled wort was determined refractometrically at 18% Brix. The cooled, hermetically sealed wort was left at room temperature (22–25 °C) for 12 months.

For LAB isolation, 5 mL of sour wort was sampled, and serial dilutions were plated on MRS agar prepared according to [23] and on acetate agar prepared according to [24]. Pure cultures were obtained by consecutive plating of serial dilutions using the pour-plate method on MRS agar, followed by transfer to liquid MRS. Cultivation was carried out at 37 °C for 24–48 hours. Culture purity was confirmed microscopically and by the absence of growth on meat-peptone agar (MPA) and Sabouraud agar.

Identification of the isolated cultures was performed by examining cell morphology and conducting standard tests used to determine affiliation with the family *Lactobacillaceae*, including catalase activity, gelatin hydrolysis, ammonia production from arginine, and carbon dioxide formation from glucose. Additionally, growth was evaluated on MRS medium supplemented with different concentrations of sodium chloride (5% and 10%) and at different initial pH values of MRS medium (4.5 and 9.0). Further phenotypic identification was carried out by assessing the isolate's ability to ferment various carbon sources: glucose, sucrose, lactose, fructose, maltose, mannose, galactose, raffinose, rhamnose, ribose, glycerol, sorbitol, inositol, mannitol, trehalose, gluconate, and D-xylose. Species identity was determined based on the obtained results by comparison with typical carbohydrate fermentation profiles described in [25] and [26]. The ABIS online service [27] was also used to determine similarity (% match) and probability (%).

Antagonistic activity was assessed according to [28] using the well-diffusion method. As opportunistic pathogens, the following strains were used: *Bacillus subtilis* UCM B-901, *Escherichia coli* UCM B-906, *Pseudomonas aeruginosa* UCM B-900, *Staphylococcus aureus* UCM B-918, and the yeast strain *Candida albicans* UCM Y-1918. The experiment was performed on MPA agar for bacterial pathogens and Sabouraud agar for yeast pathogens. Antagonistic activity was evaluated after 24 hours of incubation at 37 °C by measuring the growth inhibition zones (mm).

Antibiotic susceptibility of the culture was determined by disk diffusion on Petri dishes, according to [29]. Clinically relevant antibiotics were used [30], with the antibiotic content indicated in parentheses: ampicillin (10 μg), gentamicin (10 μg), lincomycin

(15 g), vancomycin (30 µg), oleandomycin (15 µg), nystatin (100 U), streptomycin (10 µg), benzylpenicillin (10 µg), and polymyxin B (300 U). Susceptibility was evaluated according to [31], where R denotes resistance and S denotes susceptibility.

The adhesive properties of the culture were assessed by measuring auto-aggregation and hydrophobicity. For both assays, the culture was incubated in liquid MRS medium at 37 °C for 18 hours, then centrifuged at 4000 rpm for 15 minutes. The pellet was washed twice with 0.85% NaCl solution and resuspended in the same solution to obtain an optical density of 0.3–0.4 (λ = 600 nm). Auto-aggregation was measured as described in [32], namely by incubating the prepared suspension at 37 °C for 24 hours. At 4, 6, and 24 hours, samples were collected, and optical density was measured at the same wavelength. The percentage of auto-aggregation (A) was calculated using the formula:

$$A = \left(1 - \frac{A_0 - A_t}{A_0}\right) \cdot 100\%,$$

where A_0 is the initial optical density, and A_t is the optical density after the given time interval.

Hydrophobicity of the cell surface was determined according to [33] by mixing the cell suspension with n-hexane, incubating the mixture for 30 minutes, and subsequently measuring the optical density of the aqueous phase at the same wavelength. The percentage of hydrophobicity (H) was calculated using the formula:

$$H = \frac{A_s - A_{hex}}{A_s} \cdot 100\%,$$

where A_s is the optical density of the suspension without solvent, and A_{hex} is the optical density of the suspension after incubation with n-hexane.

The following enzymatic activities were examined qualitatively: amylolytic activity according to [34], proteolytic activity according to [35], and pectinolytic activity according to [36]. All inoculated plates were incubated at 37 °C for 48 hours. Amylolytic and pectinolytic activities were evaluated by the formation of clear zones after Lugol's solution treatment; transparent zones indicated proteolytic activity.

Statistical analysis of the results. All experiments were performed in triplicate. Data are presented as mean ± standard deviation. Differences between means were assessed using Duncan's test ($P < 0.05$). Means marked with the same letters were not significantly different.

Results and Discussion

Isolates were obtained from the described nutrient media, and their number and cultural characteristics are presented in Table 1.

All isolates demonstrated similar cultural characteristics, colonies of smaller diameter were observed on acetate medium, which may be associated with an unfavorable effect of this medium on isolate growth. The negative catalase test, positive Gram staining, and the cultural characteristics presented in Table 1 indicate the potential affiliation of the isolates with LAB representatives. All selected isolates exhibited similar morphological characteristics, shown in Fig. 1, therefore, one isolate was chosen for further investigation.

According to the presented figure, short rods or coccobacilli are observed, arranged singly or in clusters, with cells exhibiting rounded ends. These morphological features indicate a potential affiliation of the isolate with the family *Lactobacillaceae*, which is characterized by cell polymorphism, including rod-shaped and cocciform forms. Other physiological and biochemical characteristics are presented in Table 2.

A negative gelatin hydrolysis result confirms the culture's affiliation with the LAB group. The presented results indicate several specific metabolic features of the investigated LAB culture, in particular its ability to grow

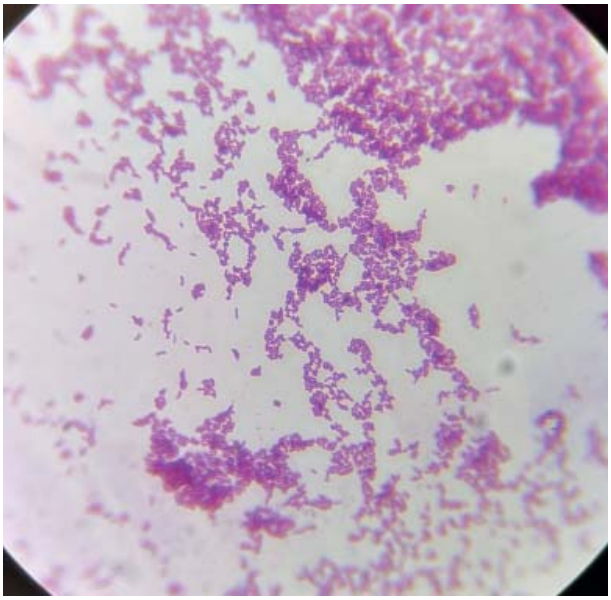

Fig. 1. **Microscopy of the selected isolate (×1000)**

Table 1. **Morphological and cultural characteristics of the isolated strains**

Nutrient medium	Number of isolates	Colony surface characteristics	Colony diameter, mm	Catalase	Gram staining
MRS	6	Round colonies with even edges, smooth, shiny surface, milky-white in color	1.5–2.0	–	+
Acetate agar	8	Round colonies, flat with a matte surface and even edges, smooth, whitish-gray in color	0.5–1.0		

Table 2. **Physiological and biochemical properties of the isolate**

Characteristic		Result
Gelatin hydrolysis		–
Arginine hydrolysis		–
Gas production from glucose		–
Growth at NaCl concentration, %	5	+
	10	+
Growth at pH	4.5	+
	9.0	+
Carbohydrate fermentation	glucose	+
	sucrose	+
	lactose	+
	fructose	+
	maltose	+
	mannitol	+
	glycerol	–
	sorbitol	+
	inositol	–
	mannose	+
	galactose	+
	raffinose	+
	rhamnose	+
	trehalose	+
	ribose	+
	gluconate	+
	D–xylose	+

at an alkaline pH (9.0) and in the presence of high NaCl concentrations (10%), suggesting osmotic tolerance. The negative results for arginine hydrolysis and gas production from glucose indicate the potential affiliation of the culture with the group of facultatively heterofermentative LAB [25]; the positive fermentation of pentoses such as ribose and D-xylose further supports this.

Based on the obtained metabolic profile and the use of the ABIS online service, the investigated culture was identified as *Lactiplantibacillus pentosus*, with a similarity of 92.6% and a probability of 98.8%. It is known that sour wort, particularly in sour beers such as lambics, is typically associated with LAB, including *Pediococcus damnosus* [37–39]. However, in [40], the authors report the presence of *Levilactobacillus brevis* and *Limosilactobacillus fermentum*. These findings do not align with the results of the present study, which may be due to differences in the raw materials and technological processes used to produce sour wort or beer. In the case of sour wort produced for sorghum beer *tchapalo*, detected LAB species included *Lpb. pentosus*, *Lpb. plantarum*, *Lvb. Brevis*, *Lmb. fermentum*, among others [21]. In [19], the authors report isolating *P. pentosaceus*, *Leuconostoc mesenteroides*, and *Lpb. plantarum* from the traditional grain-based beverage *tella*. Thus, it can be concluded that isolating LAB representatives, such as *Lpb. pentosus* and *Lpb. plantarum* is related to the initial raw materials and the specific fermentation conditions of beer wort.

Table 3. **Results of the antagonistic activity study of *Lpb. pentosus***

Opportunistic microorganism	Inhibition zone diameter, mm
B. subtilis UCM B-901	15.0 ± 0.5^{b}
E. coli UCM B-906	0.0 ± 0.0^{c}
P. aeruginosa UCM B-900	0.0 ± 0.0^{c}
S. aureus UCM B-918	30.0 ± 0.1^{a}
C. albicans UCM Y-1918	0.0 ± 0.0^{c}

The studied LAB culture *Lpb. pentosus* exhibited antagonistic activity only against *S. aureus* UCM B-918 and *B. subtilis* UCM B-901, as shown in Table 3. At the same time, the isolate did not exhibit antagonistic activity against the other tested strains of

opportunistic microorganisms: *Candida albicans* UCM Y-1918, *Pseudomonas aeruginosa* UCM B-900, and *Escherichia coli* UCM B-906.

According to source [41], *Lpb. Pentosus* demonstrated a low inhibitory effect against *Candida albicans*. However, study [42] indicates possible inhibition of *C. albicans*, with an inhibition zone diameter of 9.0 ± 0.65 mm. The absence of antimicrobial activity against *C. albicans* is explained by structural differences: bacteriocins produced by LAB act on bacterial membranes but are ineffective against fungal cells, which possess a chitin-based cell wall and ergosterol. Based on the findings reported in [43], the related species *Lpb. plantarum* exhibits vigorous inhibitory activity toward *Pseudomonas aeruginosa*, with an inhibition zone of 17.27 ± 0.04 mm, which was not observed in the present study. The low activity against *P. aeruginosa* is due to the presence of an additional outer membrane in Gram-negative bacteria, which acts as a barrier to bacteriocins and organic acids. In study [42], the authors report low antimicrobial activity of *Lpb. pentosus* against the *E. coli* strain CECT 432, with an inhibition zone diameter of 10.0 ± 1.45 mm, which is considered a low value and aligns with our findings regarding reduced antagonistic activity.

Compared with other pathogens, the present study showed high antagonistic activity against the test strains *S. aureus* and *B. subtilis*, with growth inhibition zone diameters of 30.0 ± 0.1 mm and 15.0 ± 0.5 mm, respectively. This activity is attributed to the potential synthesis of bacteriocins, which act on Gram-positive bacteria [44]. To compare the antagonistic activity of the investigated culture against *S. aureus*, literature data were used [45], where the most effective LAB strains demonstrated the following values: S6 (*Lpb. pentosus*) — 9.39 ± 0.09 mm, L19 (*Lpb. pentosus*) — 9.26 ± 0.06 mm. In comparison, the antagonistic activity of the studied isolate is 3.2 times higher. Regarding *B. subtilis*, the study [46] found no antagonism against pentocin ZFM94, a bacteriocin isolated from *Lpb. pentosus*. In study [47], the reported value is 8.10 ± 0.22 mm. The authors of [48] report antimicrobial activity of bacteriocins produced by *L. pentosus* CH2 against *B. subtilis*, with an inhibition zone of 15.0 ± 0.01 mm. These findings are consistent with those obtained in the present work.

Results of the antibiotic susceptibility tests are presented in Table. 4.

The isolated culture exhibited sensitivity only to the macrolide group (oleandomycin). Resistance to the penicillin group (ampicillin, benzylpenicillin) and glycopeptides (vancomycin) was established, which corresponds to the findings reported in studies [31, 49]. Resistance to the penicillin group may be caused by the production of β-lactamase enzymes, particularly penicillinase and cephalosporinase [50]. In the case of vancomycin resistance, which is typical for most LAB, the mechanism involves enzymes involved in D-alanine metabolism, such as D-alanine ligase, that alter the structure of the cell wall and reduce vancomycin's affinity for its target [51]. The isolate demonstrated resistance to aminoglycosides (gentamicin, streptomycin, kanamycin), which is consistent with the results described in [52]. These results are explained by the mechanisms typical of LAB, namely the

Table 4. **Antibiotic susceptibility of the *Lpb. pentosus* culture**

Group	Antibiotic	Inhibition zone, mm	Result (R/S)
Penicillins	Ampicillin	10.2 ± 0.05	R
	Benzylpenicillin	6.5 ± 0.10	R
Aminoglycosides	Gentamicin	7.0 ± 0.03	R
	Streptomycin	7.2 ± 0.08	R
	Kanamycin	8.5 ± 0.10	R
Polyenes	Nystatin	6.5 ± 0.12	R
Glycopeptides	Vancomycin	9.0 ± 0.09	R
Macrolides	Oleandomycin	15.4 ± 0.12	S
Polymyxins	Polymyxin	6.5 ± 0.05	R

Table 5. **Adhesive properties of the studied *Lpb. pentosus* culture**

Characteristic		Measured value
Auto-aggregation, %	4 h	21.81 ± 2.61
	6 h	27.31 ± 1.57
	24 h	44.80 ± 2.62
Hydrophobicity, %		46.20 ± 3.00

synthesis of enzymes (N-acetyltransferases, O-nucleotidyltransferases, O-phosphotransferases) that modify aminoglycosides. [53]. *Lpb. pentosus* exhibited resistance to polymyxin, confirming the previously established pattern of intrinsic resistance of lactobacilli to the polymyxin group of antibiotics [54]. For polyene antibiotics (nystatin), no previous studies on lactobacilli susceptibility were identified; however, according to the results obtained, resistance to this compound was observed.

The results of the auto-aggregation and cell surface hydrophobicity tests are presented in Table 5.

Lpb. pentosus demonstrated the ability to auto-aggregate with the following results: after 4 hours of incubation, the auto-aggregation level was 21.81 ± 2.61%; after 6 hours from the start of incubation, the value increased to 27.31 ± 1.57%. By the end of incubation (24 hours), the auto-aggregation level had nearly doubled compared to the 4-hour mark. In study [55], auto-aggregation values for *Lpb. pentosus* strains vary, with the highest reported value being 77.92 ± 7.22%, indicating that this trait is strain-specific. The authors of [41] report a maximum auto-aggregation level of 82.67% for *Lpb. pentosus* 68-1. For an *Lpb. pentosus* culture isolated from fermented rice, a comparable auto-aggregation value of 23% after 3 hours of incubation was reported in [56].

The mean cell-surface hydrophobicity was 46.2 ± 3.0%. Similar results were reported in a study [41], where the strain *Lpb. pentosus* 68-1, isolated from yogurt, showed a hydrophobicity level of 46.5 ± 0.38%. In contrast, for the strain *Lpb. Pentosus* MSCIN-24, isolated from the traditional fermented product mesu (fermented young bamboo shoots), the hydrophobicity level measured in *n*-hexane was only 9% [57]. This substantial difference may be associated with genetic and phenotypic characteristics of strains of different origins, particularly differences in cell-wall composition and surface proteins. The strain *L. pentosus* C22, isolated from Irish cheese, demonstrated an intermediate hydrophobicity value of 25.26% [58].

The results of enzymatic activity assessment are presented in Fig. 2.

The isolated *Lpb. pentosus* culture showed no amylolytic activity, which is confirmed by the absence of a hydrolysis zone after the addition of Lugol's solution (Fig. 2, *a*). For comparison, the strain *Lpb. pentosus* N3, isolated from the Bulgarian fermented beverage *boza*, demonstrated clear cell-associated amylolytic activity [59]. According to the results presented in Fig. 2, *b*, no proteolytic activity was observed in the studied culture. However, study [58] reports that the strain *L. pentosus* C22, isolated from Irish cheese, exhibits proteolytic activity with a clearing zone of 25 mm. At the same time, study [60] indicates that not all *L. pentosus* strains isolated from raw Tunisian milk possess proteolytic properties. The examined *Lpb. pentosus* isolate did not show any pectin hydrolysis zones (Fig. 2, *c*), indicating the absence of pectinase activity. In contrast, study [61] reports the ability of strain *Lpb. pentosus* SJ65 to synthesize pectinases.

The absence of amylolytic, proteolytic, and pectinolytic activity in the studied culture correlates with its source of isolation — acidic beer wort, which is depleted in starch, protein, and pectin. Strains exhibiting hydrolytic activities are predominantly isolated from substrates enriched in the corresponding polymers: amylolytic strains — from starch-containing substrates [59], proteolytic strains — from dairy products [58, 60], and pectinolytic strains — from plant-based materials [36, 61].

The study examined the isolation and characterization of a lactic acid bacterial culture from acidic beer wort. Based on the analysis of its morphological and physiological-biochemical properties, the isolate was identified as *Lpb. pentosus*. Antagonistic activity was demonstrated against opportunistic microorganisms, including *B. subtilis* and *S. aureus*. The antibiotic susceptibility profile indicates resistance to aminoglycosides, penicillins, and glycopeptides, typical of lactic acid bacteria. The culture exhibits moderate autoaggregation and high cell wall hydrophobicity. The absence of the enzymatic activities assessed in this work is likely attributed to the source of culture isolation. The obtained results indicate potential probiotic activity of the isolated *Lpb. pentosus*, particularly its antagonistic

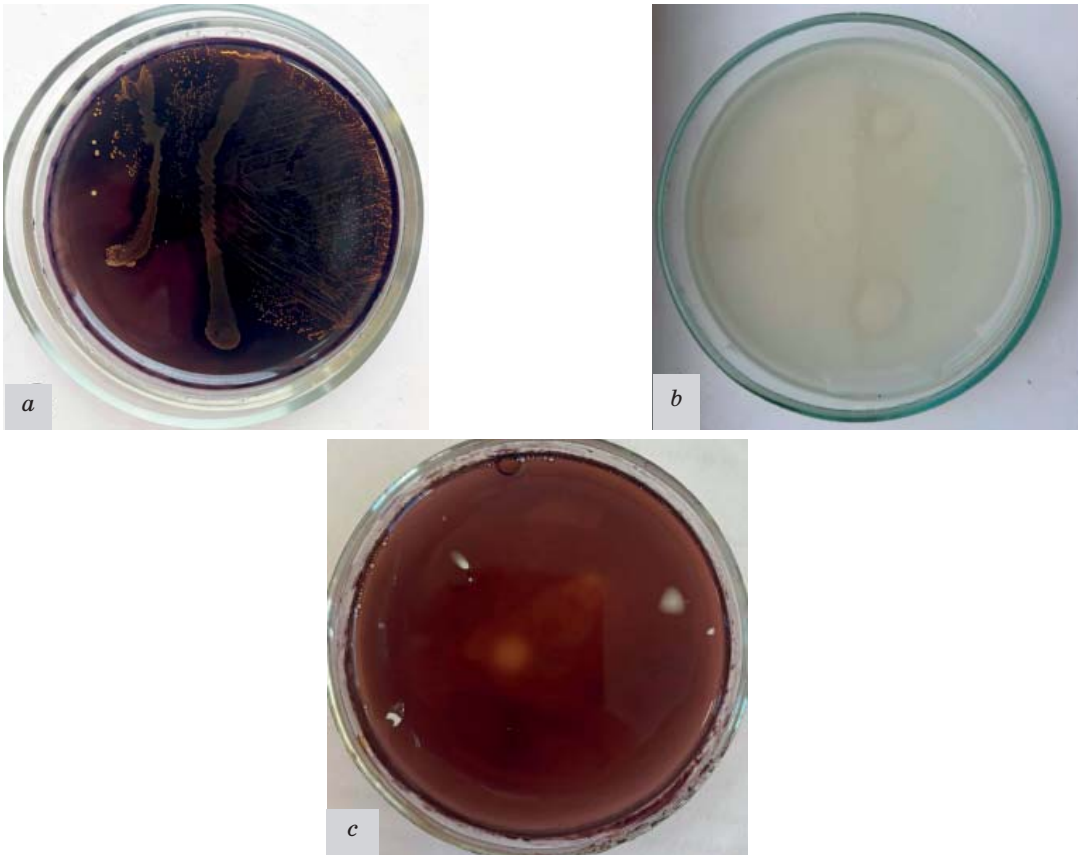

Fig. 2. **Enzymatic activities of the *Lpb. pentosus* culture:**
a — amylolytic; *b* — proteolytic; *c* — pectinolytic

action against the test culture *S. aureus*. At the same time, further studies aimed at determining the culture's stress tolerance are advisable, as they allow for conclusions regarding its ability to maintain the identified functional properties.

Authors' contribution

POS — experiment conduction, literature selection, data analysis, writing; RAI — experiment conduction, literature selection, writing; MSA — literature selection, data analysis; ZPR — writing, editing; KAD — planning, writing, editing, supervision; DSG — supervision, reviewing. All authors reviewed the manuscript and approved the submitted version.

Funding

There was no external funding.

Conflict

There was no conflict of interest.

Rerferences

1. Kieliszek, M., Pobiega, K., Piwowarek, K., Kot, A. M. (2021). Characteristics of the Proteolytic Enzymes Produced by Lactic Acid Bacteria. *Molecules,* 26(7), 1858. https://doi.org/10.3390/molecules26071858
2. Raman, J., Kim, J-S., Choi, K.R., Eun, H., Yang, D., Ko, Y-J., Kim, S-J. (2022). Application of Lactic Acid Bacteria (LAB) in Sustainable Agriculture: Advantages and Limitations. *International Journal of Molecular Sciences*, 23(14), 7784. https://doi.org/10.3390/ijms23147784
3. Garbacz, K. (2022). Anticancer activity of lactic acid bacteria. *Semin Cancer Biol.,* 86(Pt 3), 356–366. https://doi.org/10.1016/j.semcancer.2021.12.013
4. Latif, A., Shehzad, A., Niazi, S., Zahis, A., Ashraf, W., Iqbal, M., ..., Korma, S.A. (2023). Probiotics: mechanism of action, health benefits and their application in food

industries. *Front Microbiol*., 14, 1216674. https://doi.org/10.3389/fmicb.2023.1216674
5. Wang, Y., Wu, J., Lv, M., Shao, Z., Hungwe, M., Wang, J., … , Geng, W. (2021). Metabolism Characteristics of Lactic Acid Bacteria and the Expanding Applications in Food Industry. *Front Bioeng Biotechnol*., 12, 9, 612285. https://doi.org/10.3389/fbioe.2021.612285
6. Roselli M., Colafranceschi F., Cipriani V., Valle A., Zinno P., Guantario B., …, Devirgiliis, V. (2025). Isolation and Characterization of Lactic Acid Bacteria from an Italian Traditional Raw Milk Cheese: Probiotic Properties and Technological Performance of Selected Strains. *Microorganisms,* 13(6), 1368. https://doi.org/10.3390/microorganisms13061368
7. Khablenko, A., Danylenko, S., Dugan, O., Lakiychuk, O., Potemska, O. (2025). Plant-based non-alcoholic fermented beverages: microbiota overview and biotechnological production perspectives. *J. Microb. Biotech. Food Sci*., 14(6), e11295. https://doi.org/10.55251/jmbfs.11295
8. Kigel, N., Kopylova K., Danylenko, S., Naumenko, O. (2016). A study of microbiota of authentic products from different regions of Ukraine. *Food Resources*, 4(6), 212–221.
9. Barbieri, F., Tabanelli, G., Comas-Basté, O., Latorre-Moratalla, M., Angelucci, C., Gardini, F. (2025). Improvement of the safety of artisanal Spanish fermented sausages: Spotlight on the role of bacteriocinogenic *Lactiplantibacillus paraplantarum* against a Companilactobacillus alimentarius histaminogenic strain. *Food Control*, 168, 110962. https://doi.org/10.1016/j.foodcont.2024.110962
10. Danylenko, S., Kige, N., Burtseva, G. (2014). Selection of microorganisms for fermentation of meat materials. *Biotechnologia Acta*, 7(4), 107–117. https://doi.org/10.15407/biotech7.04.107
11. Stupar, J., Hoel, S., Strømseth, S., Lerfall, J., Rustad, T., Jakobse, AN. (2023). Selection of lactic acid bacteria for biopreservation of salmon products applying processing-dependent growth kinetic parameters and antimicrobial mechanisms. *Heliyon*., 9(9), e19887. https://doi.org/10.1016/j.heliyon.2023.e19887
12. Zubyk, P., Klechak, I., Dzyhun, L., Titova, L., Linovytska, V. (2025). Utilization of lignocellulosic waste from the agro-food industry by edible basidiomycetes *Pleurotus* spp. *J. Microb. Biotech. Food Sci*., 15(2). e11647. https://doi.org/10.55251/jmbfs.11647
13. Pan, X., Zhang, Y., Yue, N., Yu, K., Zhou, L., Ge, L. (2025). Isolation of Lactic Acid Bacteria from Naturally Ensiled *Rosa roxburghii* Tratt Pomace and Evaluation of Their Ensiling Potential and Antioxidant Properties. *Foods*, 14(8), 1329. https://doi.org/10.3390/foods14081329
14. Danylenko, S., Khonkov, M., Iskra, K. (2019). Lactobacilli for ensiling plant raw materials. *Agrarian Science and Food Technologies*, 5(1), 3–12.
15. Peng, C., Sun, W., Dong, X., Zhao, L., Hao, J. (2021). Isolation, identification and utilization of lactic acid bacteria from silage in a warm and humid climate area. *Sci Rep*., 11(1), 12586. https://doi.org/10.1038/s41598-021-92034-0
16. Sobrun, Y., Bhaw-Luximon, A., Jhurry, D., Puchooa, D. (2012).Isolation of lactic acid bacteria from sugar cane juice and production of lactic acid from selected improved strains. *Adv Biosci Biotechnol*., 3, 398–407. https://doi.org/10.4236/abb.2012.34057
17. Mohanan, M. M., Shetty, R., Bhat, P. S., Deepashree, V. S., Thimulappa, R. K., Bang-Berthelsen, C. H. (2025). Isolation and characterization of biological traits of millet-derived lactic acid bacteria. *Int. J. Sci. Technol*., 60(1), vvaf074. https://doi.org/10.1093/ijfood/vvaf074
18. Sornplang, P., Piyadeatsoontorn, S. (2016). Probiotic isolates from unconventional sources: a review. *J. Anim. Sci. Technol*., 19(58), 26. https://doi.org/10.1186/s40781-016-0108-2
19. Yehuala, G. A., Shibeshi, N. T., Kim, S. H., Park, M. K. (2024). Characterization of Autochthonous Lactic Acid Bacteria Isolated from a Traditional Ethiopian Beverage, Tella. *Foods*, 13(4), 575. https://doi.org/10.3390/foods13040575
20. Togo, C., Sara, S. B., Feresu, B., Mutukumira, A. (2002). Identification of Lactic Acid Bacteria Isolated from Opaque Beer (*Chibuku*) for Potential Use As a Starter Culture. *J. of Food Technol. Afr*., 7(3), 93–97. https://doi.org/10.4314/jfta.v7i3.19239
21. Solange A., Florent K. N'G., Yessé Z. N., Guillaume Y. L., André I. M., Marcellin K. D. (2010). Characterization of Lactobacillus Species Isolated from Mash, Sour Wort and Tchapalo Produced in Côte d'Ivoire. *Global Science Books*, 49–54.
22. Barth, R., Farber, M. (2019). Mastering Brewing Science: Quality and Production. Wiley & Sons, Incorporated, John. 592 p.
23. Bovo, F., Franco, L. T., Rosim, R. E., Oliveira, C. A. (2014). Ability of a Lactobacillus rhamnosus strain cultured in milk whey based medium to bind aflatoxin B1. *Food Sci Technol*., 34(3), 566–570. https://doi.org/10.1590/1678-457x.6373
24. Raccach, M. (2014). *Encyclopedia of Food Microbiology: Pediococcus*. Elsevier. 1–5.

https://doi.org/10.1016/b978-0-12-384730-0.00247-0
25. Felis, G. E., Pot, B. (2014). *The family Lactobacillaceae*. In: Lactic Acid Bacteria. Biodiversity and Taxonomy (Eds. W.H. Holzapfel, B.J.B. Wood), 245–247. https://doi.org/10.1002/9781118655252.part4
26. Whitman, W. B. (2015). *Bergey's Manual of Systematics of Archaea and Bacteria*. Wiley & Sons, Limited, John.
27. Regnum prokaryotae. ABIS online — *Lactobacillus* input. URL: https://www.tgw1916.net/bacteria_Lactobacillus_input.php (Last accessed: 03.12.2025).
28. Leska, A., Nowak, A., Szulc, J., Motyl, I., Czarnecka-Chrebelska, K. H. (2022). Antagonistic Activity of Potentially Probiotic Lactic Acid Bacteria against Honeybee (*Apis mellifera* L.) Pathogens. *Pathogens*, 16(11), 1367. https://doi.org/10.3390/pathogens11111367
29. Chowdhury, A., Choudhary, M., Sharma, V., Kant, A., Vashistt, J., Garlapati, V. K., Simal-Gandara, J. (2023). Exploration of Indian Traditional recipe "Tarvaani" from the drained rice gruel for nutritional and probiotic potential. *Int. J. Gastron. Food Sci.*, 31, 100670. https://doi.org/10.1016/j.ijgfs.2023.100670
30. Voaides, C., Boiu-Sicuia, O., Israel-Roming, F., Zamfir, M., Grosu-Tudor, S. S., Angelescu, I. R., Cornea, C. P. (2022). Lactobacillus Strains for Vegetable Juice Fermentation—Quality and Health Aspects. *Biomedicines*, 10(11), 2867. https://doi.org/10.3390/biomedicines10112867
31. Charteris, W. P., Kelly, P. M., Morelli, L., Collins, J. K. (1998). Antibiotic susceptibility of potentially probiotic *Lactobacillus* species. *J. Food Prot.*, 61(12), 1636–1643. https://doi.org/10.4315/0362-028x-61.12.1636
32. Collado, M. C., Meriluoto, J., Salminen, S. (2008). Adhesion and aggregation properties of probiotic and pathogen strains. *Eur. Food Res. Technol.*, 226, 1065–1073. https://doi.org/10.1007/s00217-007-0632-x
33. Alizadeh Behbahani, B., Noshad, M., Namazi, P., Vasiee, A. (2024). Exploring the probiotic potential of *Lactiplantibacillus pentosus* SM1: Resistance, anti-microbial activity, anti-biofilm, cytotoxic activity, and safety properties. *LWT*, 210, 116850. https://doi.org/10.1016/j.lwt.2024.116850
34. Padmavathi, T., Bhargavi, R., Priyanka, P. R., Niranjan, N. R., Pavitra, P. V. (2018). Screening of potential probiotic lactic acid bacteria and production of amylase and its partial purification. *J. Genet. Eng. Biotechnol.*, 16(2), 357–362. https://doi.org/10.1016/j.jgeb.2018.03.005
35. Ramadhan, A. R., Bachruddin, Z., Widodo, E. Y., Hanim, C. (2021). Isolation and selection of proteolytic lactic acid bacteria from colostrum of dairy cattle. *IOP Conf Ser.,* 788(1), 012077. https://doi.org/10.1088/1755-1315/788/1/012077
36. Purwanto, E. H., Frediansyah, A., Fitrianto, N., Palindung, L. S., Marwati, T., Santoso, T. J. (2024). Partial purification and pectinase activity of lactic acid bacteria and pectinolytic bacteria consortium. *IOP Conf. Ser.*, 1377(1), 012048. https://doi.org/10.1088/1755-1315/1377/1/012048
37. Straka, D., Hleba, L. (2022). Microbiological phases of spontaneously fermented beer. *J. Microb. Biotech. Food Sci.*, 12, e9624. https://doi.org/10.55251/jmbfs.9624
38. Bongaerts, D., De Roos, J., De Vuyst, L. (2021). Technological and Environmental Features Determine the Uniqueness of the Lambic Beer Microbiota and Production Process. *Appl. Environ. Microbiol.*, 26, 87(18):e0061221. https://doi.org/10.1128/AEM.00612-21
39. Spitaels, F., Wieme, A. D., Janssens, M., Aerts, M., Van Landschoot, A., De Vuyst, L. (2015). The microbial diversity of an industrially produced lambic beer shares members of a traditionally produced one and reveals a core microbiota for lambic beer fermentation. *Food Microbiol.*, 49, 23–32. https://doi.org/10.1016/j.fm.2015.01.008
40. Bokulich, N. A., Bamforth, C. W., Mills D. A. (2012). Brewhouse-resident microbiota are responsible for multi-stage fermentation of American coolship ale. *PLoS One*. 7(4), e35507. https://doi.org/10.1371/journal.pone.0035507
41. *Xue W.*, *Liu C.*, *Liu Y.*, *Ding H.*, *An C.*, *Zhang S.*, Ma, S., Zhang, Q. (2024). Probiotic Evaluation of Lactiplantibacillus pentosus 68-1, a Rutin Conversion Strain Isolated from Jiangshui, by Genomic Analysis and Tests In Vitro. *Fermentation*, 10(2), 87. https://doi.org/10.3390/fermentation10020087
42. Abriouel, H., Caballero, Gómez N., Manetsberger, J., Benomar, N. (2024). Dual effects of a bacteriocin-producing Lactiplantibacillus pentosus CF-6HA, isolated from fermented aloreña table olives, as potential probiotic and antimicrobial agent. *Heliyon*, 10(7), e28408. https://doi.org/10.1016/j.heliyon.2024.e28408
43. Li, J., Chen, X., Xie, Z., Liang, L., Li, A., Zhao, C., Wen, Y., Lou, Z. (2023). Screening and Metabolomic Analysis of Lactic Acid Bacteria-Antagonizing Pseudomonas aeruginosa. *Foods*. 12(14), 2799. https://doi.org/10.3390/foods12142799
44. Stergiou, O. S., Tegopoulos, K., Kiousi, D. E., Tsifintaris, M., Papageorgiou, A. C., Tassou, C. C., Chorianopoulos, N., Kolovos, P., Galanis, A. (2021). Whole-Genome

Sequencing, Phylogenetic and Genomic Analysis of Lactiplantibacillus pentosus L33, a Potential Probiotic Strain Isolated From Fermented Sausages. *Front Microbiol.*, 12, 746659. https://doi.org/10.3389/fmicb.2021.746659

45. Ren, D., Zhu, J., Gong, S., Liu, H., Yu, H. (2018). Antimicrobial Characteristics of Lactic Acid Bacteria Isolated from Homemade Fermented Foods. *BioMed Res. Int.*, 2018, 1–9. https://doi.org/10.1155/2018/5416725
46. Dai, M., Li, Y., Xu, L., Wu, D., Zhou, Q., Li, P., Gu, Q. (2021). A Novel Bacteriocin From *Lactobacillus Pentosus* ZFM94 and Its Antibacterial Mode of Action. *Front. Nutr.,* 8, 710862. https://doi.org/10.3389/fnut.2021.710862
47. Alizadeh, B., Jooyandeh, H., Namazi, P. (2024). The viability of Lactiplantibacillus pentosus v390 under acidic and bile conditions, and evaluation of its antimicrobial activity and safety. *Food Sci. Tech.*, 21(153), 192–206. https://doi.org/10.22034/FSCT.21.153.192
48. Mahrous, H., Mohamed, A., El-Mongy, M., El-Batal, A., Hamza, H. (2013). Study Bacteriocin Production and Optimization Using New Isolates of *Lactobacillus* spp. Isolated from Some Dairy Products under Different Culture Conditions. *Food Nutr. Sci.* 4(3), 342–356. https://doi.org/10.4236/fns.2013.43045.
49. Duche, R. T., Singh, A., Wandhare, A. G., Sangwan, V., Sihag, M. K., Nwagu, T. N. T., Panwar, H., Ezeogu, L. I. (2023). Antibiotic resistance in potential probiotic lactic acid bacteria of fermented foods and human origin from Nigeria. *BMC Microbiol.*, 23(1), 142. https://doi.org/10.1186/s12866-023-02883-0
50. *Brook, I.* (2016). Antimicrobials therapy of anaerobic infections. *J. Chemother.*, 28(3), 143–150. https://doi.org/10.1179/1973947815Y.0000000068
51. Elisha, B. G., Courvalin, P. (1995). Analysis of genes encoding D-alanine:D-alanine ligase-related enzymes in Leuconostoc mesenteroides and *Lactobacillus* spp. *Gene*, 152(1), 79–83. https://doi.org/10.1016/0378-1119(94)00692-L
52. Shao, Y., Zhang, W., Guo, H., Pan, L., Zhang, H., Sun, T. (2015). Comparative studies on antibiotic resistance in Lactobacillus casei and Lactobacillus plantarum. *Food Control.*, 50, 250–258. https://doi.org/10.1016/j.foodcont.2014.09.003
53. Zhang, Y., Zhang, N., Wang, M., Luo, M., Peng, Y., Li, Z., ..., Lu, X. (2023). The prevalence and distribution of aminoglycoside resistance genes. *Biosaf Health.*, 5(1), 14–20. https://doi.org/10.1016/j.bsheal.2023.01.001
54. Cebeci, A., Gürakan, C. (2003). Properties of potential probiotic *Lactobacillus plantarum* strains. *Food Microbiol.*, 20(5), 511–518. https://doi.org/10.1016/s0740-0020(02)00174-0
55. Montoro, B. P., Benomar, N., Lavilla Lerma, L., Castillo Gutiérrez, S., Gálvez, A., Abriouel, H. (2016). Fermented Aloreña Table Olives as a Source of Potential Probiotic *Lactobacillus pentosus* Strains. *Front Microbiol.*, 7, 1583. https://doi.org/10.3389/fmicb.2016.01583
56. Cheruvari, A., Kammara, R. (2025). Genomic Characterization and Probiotic Properties of Lactiplantibacillus pentosus Isolated from Fermented Rice. *Probiotics Antimicrob Proteins,* 17(6), 4442–4464. https://doi.org/10.1007/s12602-024-10378-1
57. Parvin, A., Adhikary, R., Guha, S., Mitra, P. K., Mandal, V. (2022). Antibiofilm and antimicrobial activity of biosurfactants from two *Lactiplantibacillus pentosus* strains against food and topical pathogens. *Journal of Food Processing and Preservation,* 46(10). https://doi.org/10.1111/jfpp.16927
58. Mohammadzadeh, M., Moayedi, A., Khomeiri, M., Zareie, Z. (2025). Exploring the probiotic properties of *Lactiplantibacillus pentosus* and gamma-aminobutyric acid production for cheese development. *Appl. Food Res.*, 100817. https://doi.org/10.1016/j.afres.2025.100817
59. Petrova, P., Emanuilova, M., Petrov, K. (2010). Amylolytic *Lactobacillus* strains from Bulgarian fermented beverage boza. *Z. Naturforsch C. J. Biosci.*, 65(3–4), 218–224. https://doi.org/10.1515/znc-2010-3-409
60. Moussa, O. B., Mankai, M., Setti, K., Bourales, M., Maher, M., Hassouna, M. (2008). Characterisation and technological properties of psychotropic lactic acid bacteria strains isolated from Tunisian raw milk. *Ann Microbiol.*, 58, 461–469. https://doi.org/10.1007/BF03175544
61. Venkatasubramanian, V., Appukuttan, S., Kadirvelu, J. (2012). Identification of pectin degrading lactic acid bacteria from fermented food sources. *Int. J. Adv. Life Sci.,* 6(1), 8–12.

ПРОБІОТИЧНІ ВЛАСТИВОСТІ МОЛОЧНОКИСЛИХ БАКТЕРІЙ ВИДІЛЕНИХ З КИСЛОГО ПИВНОГО СУСЛА

Путря О.С.[1], *Романенко А.І.*[1], *Марущенко С.А.*[1], *Зубик П.Р.*[1], *Хабленко А.Д.*[1, 2], *Даниленко С.Г.*[2]

[1]Національний технічний університет України «Київський політехнічний інститут імені Ігоря Сікорського»
[2]Інститут продовольчих ресурсів Національної академії аграрних наук України

E-mail: *khablenkoad@gmail.com*

Вивчення пробіотичного потенціалу молочнокислих бактерій, виділених із кислого пивного сусла, є перспективним напрямом у створенні нових функціональних ферментованих продуктів, оскільки такі штами можуть проявляти підвищену здатність до антагонізму та виживання за стресових умов.

Мета роботи полягала у виділенні, ідентифікації та визначенні окремих пробіотичних властивостей молочнокислих бактерій кислого пивного сусла.

Методи. Для приготування пивного сусла використовували такий метод. Виділення ізолятів молочнокислих бактерій (МКБ) проводили зі спонтанно ферментованого впродовж 12 місяців пивного сусла. Ідентифікацію проводили з використанням фізіолого-біохімічних досліджень метаболічних особливостей МКБ. Антагоністичні властивості ізоляту визначали лунковим методом, антибіотикочутливість — диск-дифузійним, адгезивні властивості — спектрофотометрично. Ферментативні активності визначали якісно.

Результати. Було ідентифіковано культуру *Lactiplantibacillus pentosus*, ізольовану з пивного сусла. Визначено її високий антагоністичний ефект по відношенню до тест-культури *S. aureus*, зона інгібування росту 30,0 ± 0,10 мм. Встановлено резистентність до аміноглікозидів, пеніцилінів, що відповідає типовому профілю антибіотикочутливості МКБ. Показники адгезії — автоагрегація та гідрофобність були 44,80 ± 2,62% та 46,20 ± 3,00 % відповідно. Досліджуваних ферментативних активностей не спостерігали.

Висновки. З кислого пивного сусла виділено *Lpb. pentosus*, що проявляє антагонізм до *S. aureus*, стійкість до аміноглікозидів, пеніцилінів та глікопептидів, середні показники автоагрегації та високу гідрофобність клітинної стінки. Виявлено відсутність досліджуваних ферментативних активностей. Результати свідчать про потенційну пробіотичну активність та потребу подальших досліджень стресостійкості.

Ключові слова: молочнокислі бактерії, кисле сусло, *Lactiplantibacillus pentosus*, антагонізм, адгезивні властивості, ферментативна активність, пробіотики, біотехнологія.

Received 2025/12/02
Revised 2026/01/14
Accepted 2026/02/12