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Optimization of Culture Conditions for Bacteriocin Production by Soil Isolates *Pediococcus pentosaceus* LB44 and *Weissella confusa* LM85

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Abstract

Background: Bacteriocins are antimicrobial proteins or peptides secreted in the culture medium during the growth of bacteria. Bacteriocin production is influenced by various physical parameters such as pH, temperature, NaCl concentration, and carbon source.

Objectives: The current study aimed at optimizing the culture conditions for higher growth and production of bacteriocins by the soil isolates, *Pediococcus pentosaceus* LB44, and *Weissella confusa* LM85.

Methods: Growth of LB44 and LM85 isolates was measured spectrophotometrically at OD_{600} and bacteriocin production was determined in terms of growth inhibition zone using agar well diffusion assay (AWDA) at different pH (3.0 to 10.0), temperatures (30 to 50°C), NaCl concentrations (2% to 10%), and different concentrations (10 to 40 g/L) of carbon sources, glucose, and lactose.

Results: *Pediococcus pentosaceus* LB44 could demonstrate similar growth and activity at pH 5.0 to 8.0; whereas *Weissella confusa* LM85 could grow and show activity at pH 5.0 to 10.0, but higher growth was recorded at pH 6.0 to 9.0. The growth and activity of *P. pentosaceus* LB44 was at 30 to 42°C, whereas *W. confusa* LM85 was able to grow and demonstrate activity at 30 to 50°C. Both isolates could grow up in 6% NaCl, but growth and activity was higher in the absence of NaCl. The isolates showed optimum growth and activity in the presence of 20 g/L glucose and 40 g/L lactose. In comparison to glucose, the growth and activity were less in the presence of lactose.

Conclusions: *P. pentosaceus* LB44 and *W. confusa* LM85 could grow and produce bacteriocin under acidic and alkaline conditions at 37°C in the medium without NaCl. These isolates utilized glucose better in comparison with lactose. The optimized conditions are helpful to recover maximum yield of bacteriocin for industrial applications.

Keywords: Antimicrobial Activity, Bacteriocin, Optimization, *Pediococcus pentosaceus*LB44, *Weissella confusa* LM85

1. Background

Lactic acid bacteria (LAB) are a group of gram-positive, acid-tolerant, generally non-sporullating rod or cocci microorganisms that produce lactic acid as the primary end product of carbohydrate fermentation (1). They are industrially important microorganisms and are used for several applications such as fermentation of dairy, meat, vegetables, production of enzymes, macromolecules, and metabolites (2). LAB is also known for the production of antimicrobial substances such as H_2O_2, CO_2, acetaldehyde, diacetyl, D-isomers of amino acids, reuterin, and bacteriocins (3). Among these, bacteriocins have promising applications in food industry as natural preservatives (4). Bacteriocins are ribosomally-synthesized extracellular peptides or proteins, which have bactericidal or static activity against other closely related species, food-borne pathogens, and spoilage bacteria. The food preservation can be achieved by applying bacteriocin or bacteriocin producing starter culture (4, 5).

Optimum growth and bacteriocin production depends on various factors such as pH, temperature, NaCl concentrations, and carbohydrate source (4, 6-10). These parameters are generally specific to producer strains (4). The growth of microorganisms depends on the rate of chemical reactions and protein structure, which are greatly influenced by temperature. Similarly, pH plays an important role in the growth of microorganisms. Variations in cytoplasmic pH inhibit the enzyme activity, and changes in external pH might cause the ionization of nutrient molecules that result in the reduction of their availability and less growth. The expression of biosynthetic genes for the production of bacteriocin may also be regulated by pH (11). Leaes et al. (12) reported that temperature and initial pH affected the expression of genes essential for the production of antimicrobial peptides. Although most of the bacteria do not require Na^+ for the growth, low NaCl concentrations can enhance the growth of LAB. The carbon source is essential for the synthesis of cell components and other metabolites (13).

Reports are available on optimization of growth conditions for many LAB, but soil isolate *Pediococcus pentosaceus* and *Weissella confusa* are least documented. The production of bacteriocins in high quantity is essentially required by food industries due to their applications. Therefore, optimization of culture conditions to produce bacteriocin is the logical extension of the authors' previous study (1). Previously, *P. pentosaceus* LB44 and *W. confusa* LM85 were isolated from soil samples collected from dairy and mulberry rhizosphere, respectively. These isolates were identified and characterized to produce bacteriocin-like inhibitory substances, which inhibited the growth of food-borne and clinical pathogens (1).

2. Objectives

The current study aimed at optimizing the culture conditions for higher growth and production of bacteriocins by soil isolates, *P. pentosaceus* LB44, and *W. confusa* LM85.

3. Methods

3.1. Bacterial Strains and Growth Conditions

P. pentosaceus LB44 and *W. confusa* LM85 were routinely grown and maintained on TGYE agar medium (g/L tryptone 5, glucose 1, yeast extract 3, agar 1.5 %, pH 7.0) at 37°C (14). MRS broth medium (g/L peptone 10, beef extract 10, yeast extract 5, dextrose 20, polysorbate-80 1, ammonium citrate 2, sodium acetate 5, magnesium sulphate 0.5, manganese sulphate 0.2, di-potassium phosphate 2, pH 6.8) (15) was used to produce bacteriocin. Indicator strain *Micrococcus luteus* MTCC 106 was grown in the nutrient broth (g/L peptic digest of animal tissue 5, NaCl 5, beef extract 1.5, yeast extract 1.5, pH 7.4). All the media components used in the current study were purchased from HiMedia, India and Sisco Research Laboratories, India.

3.2. Preparation of Cell-Free Supernatant and Determination of Antimicrobial Activity

Bacteriocins are secreted in cell-free supernatant (CFS) during the growth of bacteria. Therefore, CFS was used to determine the antimicrobial activity. The LB44 and LM85 isolates were grown in MRS medium at 37°C in a BOD incubator (Laby, India) for 18 hours. The cultures were centrifuged at 10,000 rpm for 15 minutes (Sigma, Germany). After centrifugation, CFS was collected and filter-sterilized with 0.2 μm syringe filter (Axiva, India). Agar well diffusion assay (AWDA) was performed as described by Kaur and Tiwari (1). Briefly, soft nutrient agar (0.8%) medium (5 mL) seeded with ~10^6 cells of freshly grown culture of indicator strain *M. luteus* was overlaid on nutrient base agar plate. Wells of 6 mm diameter were cut out with sterile cork borer and CFS (100 μL) was loaded. The plates were incubated at 37°C in a BOD incubator overnight. After incubation, the growth inhibition zone was observed and diameter was measured in millimeter (mm).

3.3. Optimization Conditions for Growth and Bacteriocin Production

3.3.1. pH

MRS medium was set to pH 3 to 10 with 1N NaOH and HCl. Overnight grown culture of isolate LB44 and LM85 were inoculated to an initial OD_{600} 0.02. The sets were incubated at 37°C for 18 hours. The growth was measured using a spectrophotometer (Techcomp, Hong Kong) at 600 nm and antimicrobial activity of CFS was determined using AWDA as described above.

3.3.2. Temperature

Isolate LB44 and LM85 were inoculated in MRS medium to initial OD_{600} 0.02 and incubated at 30, 37, 42, and 50°C in a BOD incubator for 18 hours. The growth was measured using a spectrophotometer at 600 nm and antimicrobial activity of CFS was determined using AWDA as described above.

3.3.3. NaCl Concentrations

MRS medium was prepared containing different concentrations of NaCl in different sets from 2% to 10%. The LB44 and LM85 isolates were inoculated to an initial OD_{600} 0.02 in each set and incubated at 37°C for 18 hours. MRS medium without NaCl was used as control. The growth and antimicrobial activity were detected as described above.

3.3.4. Different Carbon Sources

The effect of different carbon sources on growth and bacteriocin production was studied as described by Vijay Simha et al. (8). MRS medium containing different concentrations of glucose and lactose (10, 20, 30 and 40 g/L) were prepared in different sets and filter-sterilized. These sets were inoculated with LB44 and LM85 at an initial OD_{600} 0.02 followed by incubation at 37°C for 18 hours. Growth and antimicrobial activity were determined as described above. MRS medium without glucose or lactose was used as control.

3.4. Statistical Analysis

The experiments were performed in triplicate and values were expressed as mean of the 3 independent experiments with standard deviation (mean $\pm$ SD). The graphs were plotted using SigmaPlot 11.0.

4. Results

4.1. Effect of Different pH

P. pentosaceus LB44 grew up to OD_{600} 1.2 to 1.5 and demonstrated antimicrobial activity up to ~ 18 mm at pH 5.0 to 8.0. The optimum growth of these isolates was at pH 6.8 to 8.0 (OD_{600} 1.5). They grew to very less extent (OD_{600} 0.1 to 0.3) with no antimicrobial activity under highly acidic (pH 3.0 and 4.0) and alkaline (pH 9.0 and 10.0) conditions. Therefore, extreme conditions were lethal for the growth and activity. A similar pattern of growth of *W. confusa* LM85 was also observed, but it could grow under highly alkaline conditions and also demonstrated significant activity. At pH 9.0, *W. confusa* LM85 grew up to OD_{600} 1.4 and showed 17 mm growth inhibition zone. Interestingly, the isolate LM85 grew at pH 10.0 up to OD_{600} 0.7 and demonstrated 12 mm zone of growth inhibition. At pH 5.0, it grew up to OD_{600} 0.8 and showed 15 mm zone of growth inhibition (Figure 1A and 1B).

4.2. Effect of Different Temperatures

Pediococcus pentosaceus LB44 grew at 30, 37, and 42°C, but its optimum growth was recorded at 37°C (OD_{600} 1.2). The growth at 30 and 42°C were OD_{600} 1.0 and 0.7, respectively. It grew only 50% at 42°C, compared with 37°C, but LB85 isolate grew with full extent at 42°C. The LB44 Isolate did not grow at all at 50°C, whereas LM85 grew almost half with OD_{600} 0.6 at 50°C. Therefore, LB44 was sensitive to higher temperatures, whereas LM85 grew at higher temperatures up to 50°C. Antimicrobial activity demonstrated by these isolates was consistent to growth as shown in Figure 2A and 2B.

4.3. Effect of Different Concentrations of NaCl

Both of the isolates demonstrated optimum growth (OD_{600} ~ 1.3) and antimicrobial activity (the growth inhibition zone ~ 21 mm) in the medium without NaCl. The growth continuously reduced as the concentration of NaCl increased. The antimicrobial activities of these isolates also decreased by increasing the concentration of NaCl, and reduced to nil at 6% NaCl and above. These isolates could tolerate, grow, and produce bacteriocin only with up to 4% NaCl. The responses of both isolates were almost similar against different tested NaCl concentrations (Figure 3A and 3B).

4.4. Effect of Different Carbon Sources

Carbon source is important for growth and affects bacteriocin production, which is a highly energetic process. Therefore, different concentrations of the 2 most commonly used carbon sources, glucose and lactose, were supplied in the culture medium, and growth and bacteriocin production were monitored. *P. pentosaceus* LB44 grew almost equally up to OD_{600} ~ 1.6 at 10 to 40 g/L of glucose. The production rates of bacteriocin were similar (20 mm growth inhibition zone) at 20% to 40% glucose, but reduced at 10% glucose by 17 mm. *Weissella confusa* LM85 showed similar growth and bacteriocin production in the presence of different concentrations of glucose. Both isolates demonstrated much less growth (OD_{600} ~ 0.3 - 0.4), and antimicrobial activity was nil when grown in a medium without glucose (Figure 4A and 4B).

In contrast, the growth of *P. pentosaceus* LB44 was lower at 10 g/L (OD_{600} 0.4) and 20 g/L (OD_{600} 1.4) lactose, but increased with increasing the concentration of lactose. It grew optimally up to OD_{600} 1.6 at 30 to 40 g/L lactose. There was no bacteriocin production up to 20 g/L lactose used in the culture medium and very low antimicrobial activity was recorded at 30 g/L (inhibition zone ~ 9 mm). The optimum activity (growth inhibition zone ~16 mm) was recorded at 40 g/L lactose. Almost similar patterns of growth and bacteriocin production were also demonstrated by *W. confusa* LM85 (Figure 5A and 5B). In comparison to glucose, growth and bacteriocin production by both isolates were lower at similar concentrations of lactose. Both isolates demonstrated bacteriocin production at 10% to 20% glucose, but it was nil in the presence of lactose at these concentrations. These results suggested that both isolates utilized glucose more efficiently than lactose.

5. Discussion

The production of bacteriocin is an energetic process, which show growth associated phenomenon. Different conditions including carbon source are important for the production of bacteriocin. It is reported that antimicrobial activity follows similar patterns of growth curve of most LAB strains (14, 16, 17). Therefore, growth was optimized under different conditions such as pH, temperature, different concentrations of NaCl and carbon source. The LB44 Isolate grew and demonstrated activity at acidic and near neutral pH, whereas LM85 demonstrated growth and activity at acidic, near neutral, and highly alkaline pH. *Lactobacillus casei* and *Lactococuss lactis* subsp. *lactis* showed optimum growth and bacteriocin production at pH 6.5 to 7.0 (4). *Pediococcus pentosaceus* MTCC 5151 showed optimum activity at pH 5.5 (18). *pentosaceus* NRC AM1 and *P. pentosaceus* NRC AM4 grew well at pH 4.0 to 8.0 (19).

P. pentosaceus LB44 and *W. confusa* LM85 optimally grew and demonstrated activity at 30 to 37°C. The growth and activity of LB44 was almost half, compared with LM85 at 42°C. *Pediococcus pentosaceus*LB44 did not grow or show activity

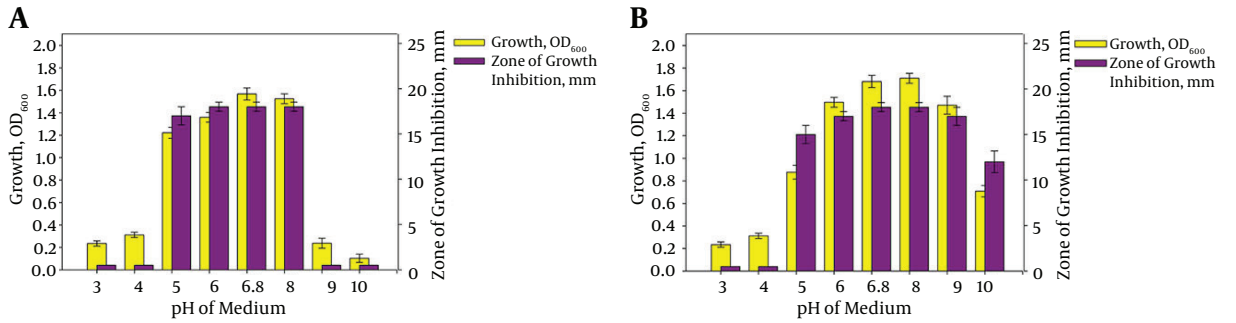

Figure 1. Growth and Antimicrobial Activity of Isolates LB44 (A) and LM85 (B) at Different pH

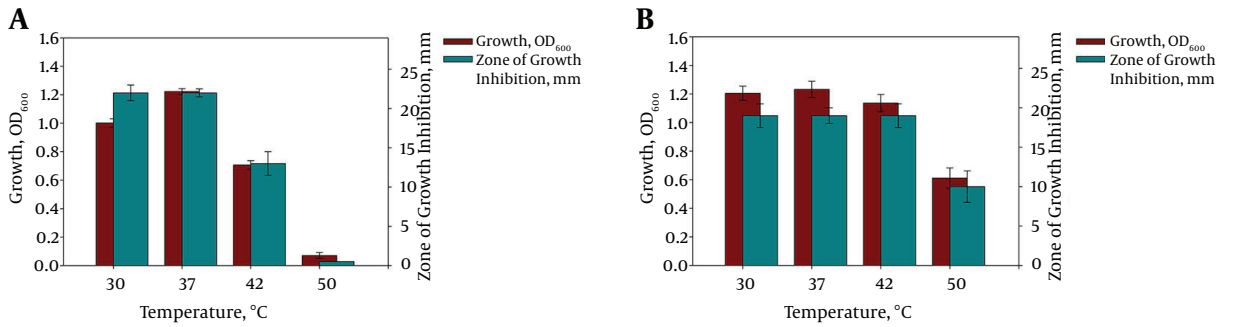

Figure 2. Growth and Antimicrobial Activity of Isolates LB44 (A) and LM85 (B) at Different Temperatures

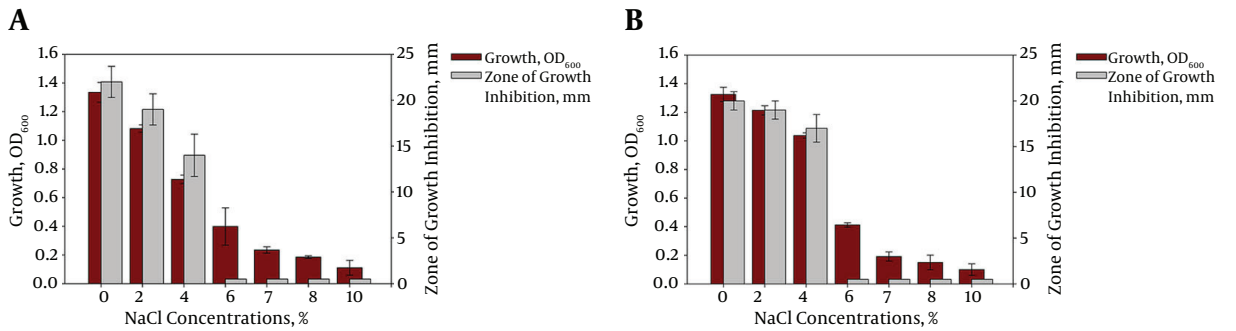

Figure 3. Growth and Antimicrobial Activity of Isolates LB44 (A) and LM85 (B) at Different Concentrations of NaCl

at 50°C, but *W. confusa* LM85 grew and demonstrated activity up to half compared to 37°C. The optimum temperature for the growth of *P. pentosaceus* strains was 28 to 35°C (20). *P. pentosaceus* NRC AM1 and *P. pentosaceus* NRC AM4 grew well at 10 to 45°C (19). The optimum temperatures for the production of pediocin by *P. pentosaceus* ST18 and *P. pentosaceus* ACCEL were 30 and 37°C, respectively (21, 22). The maximum bacteriocin production by *P. acidilactici* 13 and *P. pentosaceous* NCDC 273 was reported at 37°C (8, 13). Optimum bacteriocin production by *L. casei* LA-1 was reported at 26 to 37°C (4). *Weissella cibaria* grew at 20 to 37°C, but there was no growth at 45°C (OD_{600} 0.169). The optimum growth and activity of *W. cibaria* 110 was reported at 30°C (23). Pal et al. (17) reported bacteriocin production by *W. paramesenteroides* DFR-8 at 25 to 40°C, but optimum activity was at 35°C. The optimum production of bacteriocin by *Lactococcus lactis* subsp. *lactis* A164 and *Lactobacillus rhamnosus* GP1 were reported at pH 6.0 and 30°C (16, 24). The soil isolate *W. confusa* LM85 grew at alkaline pH 9 and 10, and higher temperature up to 50°C, which was a unique fea-

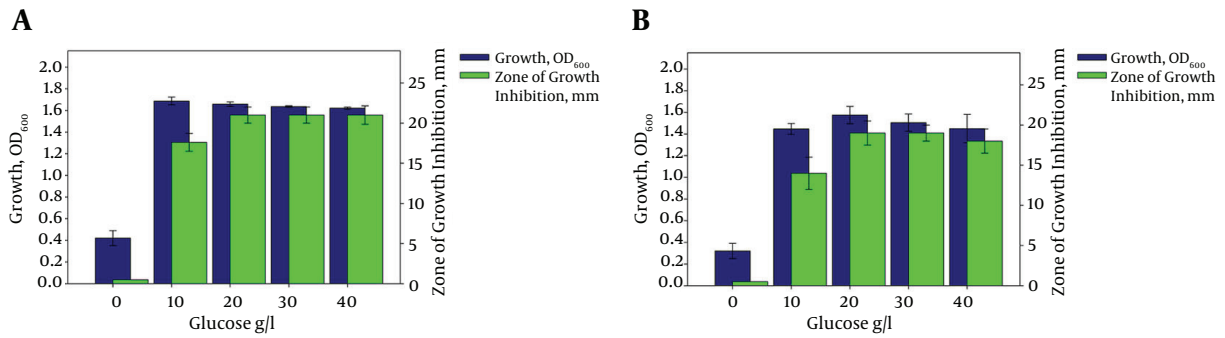

Figure 4. Growth and Antimicrobial Activity of Isolates LB44 (A) and LM85 (B) at Different Concentrations of Glucose

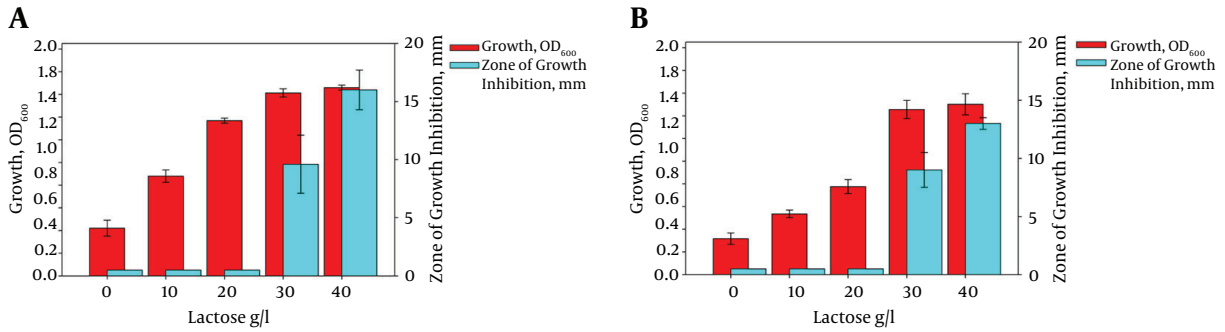

Figure 5. Growth and Antimicrobial Activity of Isolates LB44 (A) and LM85 (B) at Different Concentrations of Lactose

ture and not reported till date, to the best of the authors' knowledge. The growth at broader range of pH and temperature provides extra advantage for the application of isolates and their bacteriocins in different foods.

Growth of both isolates decreased with increase in NaCl concentrations. Similar results were also reported for *P. pentosaceus* NRC AM4 where growth decreased with increase in NaCl concentrations (19). *Pediococcus acidilactici* 13 grew up to 10% NaCl, but grew optimally in the absence of NaCl (13). The growth of LAB is sometimes better in the presence of low salt concentration, usually 1% to 2% and is inhibited above 3% NaCl, while few LAB are more resistant to NaCl (13). Delgado et al. (6) stated that NaCl was required to maintain osmotic pressure in the cells, but not required for the production of bacteriocin.

Both isolates grew optimally and demonstrated activity in 10% to 20% glucose. The growth and activity were less in the presence of lactose at similar concentrations. Similar observation was reported by Pal et al. (17) where optimum bacteriocin production by *W. paramesenteroides* DFR-8 was higher in the presence of glucose as compared to those of other carbon sources including lactose. Delay in growth and antimicrobial activity by *P. pentosaceus* NCDC 273 in the presence of lactose was also reported by Vijay Simha et al. (8). These findings suggested that the growth and bacteriocin production were higher in the presence of glucose as compared to that of lactose. This could be due to higher utilization of glucose in comparison with that of lactose during the growth of LB44 and LM85 isolates. The optimized condition for growth and bacteriocin production by these isolates are helpful to recover higher yield for their possible applications.

5.1. Conclusions

The culture conditions were optimized for higher growth and production of bacteriocin by soil isolates *P. pentosaceus* LB44 and *W. confusa* LM85. The optimum growth conditions for bacteriocin production by isolate LB44 was MRS medium supplemented with glucose at pH 6.8 and incubation temperature 37°C. The LM85 isolate grew in highly alkaline conditions and higher temperatures, but LB44 failed to grow and produce bacteriocin under such conditions. The growth and activity of both isolates were higher in the medium without NaCl. Glucose was more effective than lactose for growth and bacteriocin production. Therefore, optimized culture conditions are

useful for higher yield of bacteriocins and their industrial applications.

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Footnote

Authors' Contribution: All authors had equal contribution in design of work, data interpretation, and manuscript writing.

References

1. Kaur R, Tiwari SK. Isolation, identification and characterization of Pediococcus pentosaceus LB44 and Weissella confusa LM85 for the presence of bacteriocin-like inhibitory substances (BLIS). *Microbiology.* 2016;**85**(5):540–7. doi: 10.1134/s0026261716050088.
2. Aidani E, Delfan P, Akbarian M, Akbarian A, Ghasemkhani N, Moayedi F. Genomics of Lactic Acid Bacteria: Features, Function, and Comparative genomics: A review. *Int J Biosci.* 2014;**5**(4):34–46. doi: 10.12692/ijb/5.4.34-46.
3. Gupta A, Tiwari SK. Probiotic Potential of Lactobacillus plantarum LD1 Isolated from Batter of Dosa, a South Indian Fermented Food. *Probiotics Antimicrob Proteins.* 2014;**6**(2):73–81. doi: 10.1007/s12602-014-9158-2. [PubMed: 24676724].
4. Kumar M, Jain AK, Ghosh M, Ganguli A. Statistical optimization of physical parameters for enhanced bacteriocin production by L. casei. *Biotechnol Bioproc Engin.* 2012;**17**(3):606–16. doi: 10.1007/s12257-011-0631-4.
5. Mekri M, Abbouni B, Tifrit A, Daouadji KL, Doumandji A. Antibacterial activity of lactic acid bacteria isolated from raw goat's milk against spoilage and pathogenic bacteria. *Global Veterinaria.* 2015;**15**(5):485–92.
6. Delgado A, Arroyo Lopez FN, Brito D, Peres C, Fevereiro P, Garrido-Fernandez A. Optimum bacteriocin production by Lactobacillus plantarum 17.2b requires absence of NaCl and apparently follows a mixed metabolite kinetics. *J Biotechnol.* 2007;**130**(2):193–201. doi: 10.1016/j.jbiotec.2007.01.041. [PubMed: 17462774].
7. Bing H, Zhanqiao Y, Baosheng L, Qingshan M, Rijun Z. Optimization of bacteriocin production by Lactobacillus plantarum YJG, isolated from the mucosa of the gut of healthy chickens. *Afr J Microbiol Res.* 2011;**5**(10):1147–55. doi: 10.5897/ajmr10.643.
8. Vijay Simha B, Sood SK, Kumariya R, Garsa AK. Simple and rapid purification of pediocin PA-1 from Pediococcus pentosaceous NCDC 273 suitable for industrial application. *Microbiol Res.* 2012;**167**(9):544–9. doi: 10.1016/j.micres.2012.01.001. [PubMed: 22277956].
9. Garsa AK, Kumariya R, Sood SK, Kumar A, Kapila S. Bacteriocin production and different strategies for their recovery and purification. *Probiotics Antimicrob Proteins.* 2014;**6**(1):47–58. doi: 10.1007/s12602-013-9153-z. [PubMed: 24676767].
10. Bali V, Panesar PS, Bera MB, Kennedy JF. Bacteriocins: Recent Trends and Potential Applications. *Crit Rev Food Sci Nutr.* 2016;**56**(5):817–34. doi: 10.1080/10408398.2012.729231. [PubMed: 25117970].
11. Motta AS, Brandelli A. Evaluation of environmental conditions for production of bacteriocin-like substance by Bacillus sp. strain P34. *World J Microbiol Biotechnol.* 2007;**24**(5):641–6. doi: 10.1007/s11274-007-9520-6.
12. Leaes FL, Velho RV, Caldas DG, Pinto JV, Tsai SM, Brandelli A. Influence of pH and temperature on the expression of sboA and ituD genes in Bacillus sp. P11. *Antonie Van Leeuwenhoek.* 2013;**104**(1):149–54. doi: 10.1007/s10482-013-9935-z. [PubMed: 23677688].
13. Altuntas EG, Cosansu S, Ayhan K. Some growth parameters and antimicrobial activity of a bacteriocin-producing strain Pediococcus acidilactici 13. *Int J Food Microbiol.* 2010;**141**(1-2):28–31. doi: 10.1016/j.ijfoodmicro.2010.04.024. [PubMed: 20483184].
14. Tiwari SK, Srivastava S. Purification and characterization of plantaricin LR14: a novel bacteriocin produced by Lactobacillus plantarum LR/14. *Appl Microbiol Biotechnol.* 2008;**79**(5):759–67. doi: 10.1007/s00253-008-1482-6. [PubMed: 18496687].
15. De Man JC, Rogosa M, Sharpe ME. A Medium for the Cultivation of Lactobacilli. *J Appl Bacteriol.* 1960;**23**(1):130–5. doi: 10.1111/j.1365-2672.1960.tb00188.x.
16. Cheigh CI, Choi HJ, Park H, Kim SB, Kook MC, Kim TS, et al. Influence of growth conditions on the production of a nisin-like bacteriocin by Lactococcus lactis subsp. lactis A164 isolated from kimchi. *J Biotechnol.* 2002;**95**(3):225–35. [PubMed: 12007863].
17. Pal A, Ramana KV, Bawa AS. Simplification and optimization of deMan Rogosa Sharpe (MRS) medium for enhanced production of bacteriocin by Weissella paramesenteroides DFR-8. *J Food Sci Technol.* 2010;**47**(3):258–65. doi: 10.1007/s13197-010-0040-2. [PubMed: 23572634].
18. Agrawal R, Dharmesh S. An anti-Shigella dysenteriae bacteriocin from Pediococcus pentosaceus MTCC 5151 cheese isolate. *Turk J Biol.* 2012;**36**(2):177–85.
19. Mabrouk AMM, Effat BAM, Sadek ZIM, Tawfiek NF, Hassan ZMR, Magdoub MNI. Antibacterial activity of some Lactic acid Bacteria isolated from Egyptian Dairy Products. *Int J Chem Tech Res.* 2014;**6**(2):1139–50.
20. Papagianni M, Anastasiadou S. Pediocins: The bacteriocins of Pediococci. Sources, production, properties and applications. *Microb Cell Fact.* 2009;**8**:3. doi: 10.1186/1475-2859-8-3. [PubMed: 19133115].
21. Wu CW, Yin LJ, Jiang ST. Purification and characterization of bacteriocin from Pediococcus pentosaceus ACCEL. *J Agric Food Chem.* 2004;**52**(5):1146–51. doi: 10.1021/jf035100d. [PubMed: 14995112].
22. Todorov SD, Dicks LMT. Pediocin ST18, an antilisterial bacteriocin produced by Pediococcus pentosaceus ST18 isolated from Boza, a traditional cereal beverage from Bulgaria. *Process Biochem.* 2005;**40**:365–70.
23. Srionnual S, Yanagida F, Lin LH, Hsiao KN, Chen YS. Weissellicin 110, a newly discovered bacteriocin from Weissella cibaria 110, isolated from plaa-som, a fermented fish product from Thailand. *Appl Environ Microbiol.* 2007;**73**(7):2247–50. doi: 10.1128/AEM.02484-06. [PubMed: 17293526].
24. Sarika AR, Lipton AP, Aishwarya MS. Bacteriocin production by a new isolate of Lactobacillus rhamnosus GP1 under different culture conditions. *Adv J Food Sci Technol.* 2010;**2**(5):291–7.