

March 27th and 28th, 2025 27 e 28 de Março, 2025 WYNDHAM SÃO PAULO IBIRAPUERA CONVENTION PLAZA SÃO PAULO - BRAZIL

Enterococcus spp. are predominate bacteriocinogenic strains in Feta Cheese, however, Pedicoccus acidilactici can be the best candidate for the further biopreservation

Vitor Luis Fagundes^{1,2}, Marcos Vinício Alves², Julia Arantes Galvão¹, Svetolsav Dimitrov Todorov^{2,3}

^{1.} UFPR, Universidade Federal do Paraná, Curitiba, 80035-050, PR, Brasil;

^{2.} USP, Universidade de São Paulo, São Paulo, 05508-000, SP, Brasil;

^{3.} IPVC, Instituto Politécnico de Viana do Castelo, 4900-347 Viana do Castelo, Portugal;

Introduction: Lactic Acid Bacteria (LAB) and their metabolites are promising alternatives to promote food safety, quality assurance and even serve as probiotics and postbiotics. Moreover, food borne pathogens such as Campylobacter jejuni, Listeria monocytogenes, Salmonella enterica, Escherichia coli and *Staphylococcus aureus* are some of the most frequent food safety concerns. The crossing point between beneficial and spoilage are some of the metabolites produced by LAB, representing antagonistic activity and their applications as biopreservatives, meeting the growing consumer demand for alternatives to chemical preservatives (Choi et al., 2023).

Objective: The aim of this study was to isolate bacteriocin-producing LAB from traditional fermented food products and evaluate their potential as putative food protective agents.

Results: In the preliminary screening LAB from different samples of dairy origin were evaluated for the bacteriocin production properties. A set of 14 preliminary identified as LAB based on catalase test and Gram staining, were isolated from Bulgarian feta cheese. After the confirmation that production antimicrobials by selected LAB were on proteinaceous nature (bacteriocin/s), the strains were differentiated by repPCR and identified based on recommendations from Burgey's Manual (de Vos et al., 2009) and following 16S rRNA partial gene sequencing. Tested 14 isolates were grouped in 5 different clusters and representative from each groups identified as Enterococcus faecium (K33), Enterococcus faecalis (K35, K37, K40) and Pediococcus acidilactici (K41). The spectrum of activity for the selected 5 strains was evaluated against taxonomically related microorganisms, including different Listeria monocytogenes strains and several beneficial strains from the microbial collection of ProBacLab, Laboratory of Food Microbiology, Faculty of Pharmaceutical Sciences, University of Sao Paulo, Sao Paulo, Brazil. All tested strains represented strong anti-Listerial activity. The studied bacteriocins were shown to be stable to variations in pH, temperature and against chemical compounds used in the food industry (Tween 20 and 80, NaCl, milk and SDS). Selected 5 strains were presented reasonable survive in simulated in vitro conditions of the gastrointestinal system. Moreover, related to the safety assessment experiments, the studied LAB strains demonstrated safety profile, regarding the hemolytic outline and proteolytic and gelatinase activity, which were not detected. The antibiotic resistance/susceptibility based on physiological and biomolecular, and presence of virulence genes were also assessed. It was interesting that two of the isolates exhibited resistance to vancomycin (Enterococcus faecalis K40 and Pediococcus acidilactici K41) and potential virulence factors were found in some strains, namely gel (in all), esp (in Enterococcus faecalis K40), efa and asa (in Enterococcus faecium K33 and Enterococcus faecalis K35, K37 and K40).



March 27th and 28th, 2025 27 e 28 de Março, 2025 WYNDHAM SÃO PAULO IBIRAPUERA CONVENTION PLAZA SÃO PAULO - BRAZIL

Conclusion: Compared with the other isolates, Pediococcus acidilactici K41 proved to be a good candidate for use as an agent to be incorporated as a biopreservative. However, although the other isolates possess virulence genes, the bacteriocins produced by them can be incorporated, after purification, in food production to prevent the growth of potentially pathogenic microorganisms, such as Listeria.

References

De Vos, P., Garrity, G.M., Jones, D., Krieg, N.R., Ludwig, W., Rainey, F.A., Schleifer, K.-H., & Whitman, W.B. 2009. Bergeys manual of systematic bacteriology: the firmicutes. London: Springer.

Choi GH, Holzapfel WH, Todorov SD. Diversity of the bacteriocins, their classification and potential applications in combat of antibiotic resistant and clinically relevant pathogens. Crit Rev Microbiol. 2023 Sep;49(5):578-597. doi: 10.1080/1040841X.2022.2

Acknowledgements: FCT (UIDB/05937/2020 and UIDP/05937/2020), FAPESP (2023/05394-9; 2023/14944-2), CAPES (88887.977258/2024-00), CAPES (88887.941664/2024-00).