SCIENTIFIC OPINION

by Assoc. Prof. Elena Bozhidarova Krachmarova, PhD

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Regarding dissertation entitled “Novel Producers of Prebiotic Carbohydrates from Bulgarian Yoghurt”, submitted for the acquisition of the educational and scientific degree “Doctor” by the full-time PhD candidate Ivan Dimitrov Ivanov in Filed of Higher Education 4.3. Biological Sciences, Scientific Specialty Microbiology, at the Institute of Microbiology “Stefan Angelov” – BAS.

Supervisors: Prof. Penka Mladenova Petrova, DSc (IMicB – BAS) and Prof. Kaloyan Kirilov Petrov, DSc (IChH – BAS).

1. General Information on the Dissertation and the Doctoral Candidate

The dissertation by Ivan Dimitrov Ivanov has been submitted in accordance with the Law on the Development of the Academic Staff in the Republic of Bulgaria and the Regulations for its implementation. The dissertation fully meets the Regulations for the Conditions and Procedure for Acquiring Academic Degrees and Holding Academic Positions at the Institute of Microbiology "Stefan Angelov" at the Bulgarian Academy of Sciences.

The dissertation is 121 pages long and includes 27 figures, 13 tables, and 218 bibliographic references. The submitted materials also include the PhD candidate’s CV; a list of publications related to the dissertation and their citations; conference participations; certificates from specialized courses; and an abstract (in Bulgarian and English).

2. Relevance and Significance of the Topic

The dissertation is dedicated to establishing the foundations of a biotechnological process to produce galactooligosaccharides (GOS). It provides an analysis of the role of lactic acid bacteria (LAB) in traditional and modern fermented foods, their probiotic potential, and their capacity to synthesize bioactive compounds that support beneficial gastrointestinal microbiota. The topic is highly relevant due to the increasing scientific and industrial interest in functional foods and the role of prebiotics in maintaining human microbiome health.

3. Objectives, Tasks, and Research Methodology

The main objective of the dissertation is the molecular characterization of the lacZ gene and β-galactosidase from a Bulgarian strain of Lactobacillus delbrueckii ssp. bulgaricus, with the aim of optimizing the in vitro synthesis of prebiotic GOS. The research tasks are fully aligned with the stated objective. The microbiological, analytical, and molecular-biological methods employed are appropriate and sufficient for the successful completion of the research tasks.

4. Main Scientific Results and Contributions

The principal contributions include:
1. First metagenomic profiling of homemade Bulgarian yoghurts from diverse geographical regions, revealing both dominant and rare/new LAB species.
2. First evidence for the presence of L. equicursoris in yoghurt.
3. Identification of L. bulgaricus strain 43 with exceptionally high transgalactosidase activity.
4. Sequencing and characterization of the strain's lacZ gene, revealing 21 unique amino acid substitutions stabilizing the tetrameric structure and leading to markedly enhanced enzymatic activity.
5. Successful heterologous expression of recombinant β-galactosidase, demonstrating significant industrial potential for GOS production with prebiotic properties.

5. Evaluation of the Publications Related to the Dissertation

The dissertation results have been published in three scientific articles: one in a Q1 journal (International Journal of Molecular Sciences) and two in Q2 journals (Processes and Microorganisms), according to Clarivate Journal Citation Reports. The candidate's publications have been cited 100 times (excluding self-citations). These metrics attest to the high scientific value and contemporary relevance of the research.

6. Conclusion

After reviewing the dissertation, abstract, and accompanying materials, and based on the analysis of their significance and the scientific and applied contributions presented, I confirm that the dissertation meets the requirements of the LDASRB, its Implementing Regulations, and the BAS Regulations, as well as the criteria of the Institute of Microbiology "Stefan Angelov", BAS, for awarding the degree Doctor. The candidate meets the minimum national requirements for the professional field.

I give my positive evaluation of the dissertation and recommend that the esteemed scientific jury award the educational and scientific degree Doctor to Ivan Dimitrov Ivanov in Professional Field 4.3. Biological Sciences, Scientific Specialty Microbiology.

18 November 2025

Prepared by:
Assoc. Prof. Elena Bozhidarova Krachmarova, PhD
(Member of the Scientific Jury)