



БИОЛОГИЧЕСКИ ФАКУЛТЕТ
СОФИЙСКИ УНИВЕРСИТЕТ



FACULTY OF BIOLOGY
SOFIA UNIVERSITY

SCIENTIFIC STATEMENT

on a dissertation work for obtaining the educational and scientific degree "**doctor**" in the field of higher education 4. Natural sciences, mathematics and informatics, professional direction 4.3. "Biological Sciences", PhD program "Microbiology"

Author of the dissertation: Milena Nikolova Petrova, PhD student at the Department General and Industrial Microbiology in the Faculty of Biology of Sofia University "St. Kliment Ohridski"

Topic of the dissertation: Development of biologically active products from new natural sources

Member of the Scientific Jury: Miroslava Konstantinova Zhiponova, PhD, Associate Professor, Sofia University "St. Kliment Ohridski", Faculty of Biology, Department of Plant Physiology, scientific specialty "Plant Physiology", member of the SJ appointed by order of the Rector of SU № РД-38-389/13.07.2022

1. Relevance of the problem.

In the dissertation is applied an approach to test alternative antibiotics based on bioactive substances from new natural sources, which include products from microalgae, natural antibiotics isolated from actinobacteria and hemocyanin from marine crabs. Such a strategy is relevant to suppress the existing trends of emerging resistance towards widespread used antibiotics, as well as against the application of pesticides polluting the environment. The newly researched products with proven antimicrobial and antioxidant properties would have applications in the treatment of foodborne diseases, for food storage and in agriculture against phytopathogens.

2. Degree of knowledge of the state of the problem and creative interpretation of the literature review.

Milena Petrova is very familiar with the theory and the available in the literature data related to the dissertation topic, which is reflected in the literature review and the cited over 500 references. A detailed introduction to natural sources of antimicrobial substances is presented, including general characteristics of microalgae and specific information about the species studied in the dissertation. Additional sources of antimicrobial substances, such as hemocyanin-synthesizing arthropods and molluscs, higher plants, and microorganisms, have also been examined. Pathogenic bacteria are defined, and characteristics of studied phytopathogenic bacteria species and commercially available biological control products are described in detail. Human pathogens included in the work are also described. Methods for introducing a natural sample into laboratory conditions are presented. A synergistic effect between microalgae and other antibiotic products has been reviewed.

3. Purpose, tasks, hypotheses and research methods. Correspondence of the chosen research methodology with the set goal and tasks of the dissertation work.

The aim of the dissertation is the research of new potential antibiotics of natural origin. The tasks include characterization of promising microalgae strains and analysis of antioxidant and antimicrobial indicators of microalgae products (extracts, fractions, extracellular compartments). Additional tasks include determining the antimicrobial activity of a combination of microalgae extracts, natural antibiotics isolated from actinobacteria, and hemocyanin from marine crabs.

In accordance with the set goal and tasks, a wide range of material (microalgae and microorganisms) and methods (molecular, biochemical, antibacterial, mathematical) have been selected and described in detail. The added appendices further assist to guide and track the individual steps of the multiple set of studies. The PhD student has explored details in each of the experimental approaches used and has presented them quite clearly and comprehensibly.

4. Representation and interpretation of the obtained results.

Milena Petrova grouped the results according to the applied methodology into molecular, biochemical, antioxidant and antibacterial. It is impressive the diligent shaping of the results and their summarization as tables, which facilitates their perception: a total of over 50 (41+A11) tables and over 50 (26+A28) figures. Sufficient repetitions were performed to support the statistical validity of the results.

5. Discussion of the results and used references.

The PhD student interprets the results thoroughly, including a comparison with Bulgarian and foreign references. In total, 11 conclusions and 5 contributions are drawn, which highlight that microalgal extracts have promising potential for the development of antimicrobial agents.

6. Dissertation contributions.

Fundamental, methodical and applied contributions of the dissertation related to new natural sources of bioactive products are highlighted. Taxonomic, biochemical and antimicrobial characteristics of poorly studied microalgae strains were investigated in detail. The resulting variants of microalgae extracts have potential antioxidant activity and action against a wide range of human pathogens and phytopathogens. Software has been developed to efficiently estimate zones of inhibition.

7. Assessment of the degree of personal involvement of the PhD student in the contributions.

For the preparation of the dissertation, Milena Petrova has acquired methodological skills, with which she has contributed to their development and effective application, especially in the field of microbiology. The careful preparation of the dissertation with a detailed comparative analysis of the information available in the literature demonstrates a strong motivation, a thorough and responsible attitude to the research goals and tasks. My assessment of the PhD student's personal contribution is excellent.

My personal impressions of the PhD student are also positive – Milena is a purposeful young person who, with attention, diligence and persistence, achieves results that she successfully analyzes and interprets.

8. Critical notes and questions

I would recommend Milena Petrova to work in the direction of real practical application of the new natural alternatives to antibiotics she has researched.

9. Published articles and citations

Three articles on the dissertation have been published, two of them in specialized refereed journals (SJR) and in journals with an impact factor (IF). The publications indicate the PhD student's ability to present her scientific achievements at international level, which is a prerequisite for the recognition of her works by other authors. The presented dissertation summary reflects objectively the structure and content of the dissertation work.

The work in the dissertation is supported by two research projects financed by the Scientific Research Fund of Sofia University "St. Kliment Ohridski", which testifies to responsibility and ability to attract funds to finance the research work.

CONCLUSION:

Based on the applied by the PhD student various research methods, the correctly conducted experiments, the performed summaries and conclusions, I consider that the presented dissertation meets the requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for its implementation and the respective Regulations in the Faculty of Biology and SU, which gives me reason to evaluate it **POSITIVELY**.

I propose to the honorable Scientific Jury to vote positively, as well, and to award Milena Nikolova Petrova the educational and scientific degree "**doctor**" in the professional direction 4.3. Biological Sciences, PhD Program "Microbiology".

Date: 08.09.2022

Member of the Scientific Jury:

Sofia

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(Miroslava Zhiponova)