#### **SCIENTIFIC OPINION**

## by Assoc. Prof. RADOSLAV IGNATOV ABRASHEV, PhD

The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences

On the materials submitted for participation in the competition for the academic position of "ASSOCIATE PROFESSOR" in professional field 4.3. Biological Sciences, scientific specialty Microbiology

By order No. I-77 / 28.05.2025 of the Director of the Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences (SAIM-BAS), I am included in the Scientific Jury for the selection of "Associate Professor" for the needs of the Laboratory "Extremophilic Microorganisms" at the Department of General Microbiology at SAIM-BAS.

In the competition for "Associate Professor", announced in the State Gazette, issue 110/31.12.2024, documents have been submitted by Dr. Ivanka Petrova Boyadzhieva, Chief Assistant Professor at the Laboratory of Extremophilic Microorganisms, Department of General Microbiology at the SAIM-BAS. To participate in the competition, she has submitted the necessary documents and materials proving the fulfillment of the requirements for holding the academic position of Associate Professor on an electronic medium. All of them are in accordance with the Law on the Development of the Academic Staff in the Republic of Bulgaria (LDASRB), the Regulations for its implementation, as well as the Regulations of the SAIM-BAS. The competition documentation is well-written and comprehensively reflects the candidate's research activities.

#### CAREER DEVELOPMENT OF THE CANDIDATE

Dr. Ivanka Petrova Boyadzhieva holds a master's degree in molecular biology, specialization Virology from Sofia University "St. Kliment Ohridski", Faculty of Biology since 1999. Immediately after that, in 2000, she joined the Institute of Microbiology in the Laboratory "Extremophilic Microorganisms". Since 2011, she has been an assistant, and since 2015, a chief assistant in the same laboratory. In 2008, she received the educational and scientific degree "Doctor" at the SAIM-BAS. The candidate's work experience is more than 25 years.

For 2 years (from 2023 to the present), Dr. Boyadzhieva has also performed administrative duties as head of the Laboratory "Extremophilic Microorganisms" at the Institute.

The candidate's scientific and research activities are entirely related to the topic of the competition and reflect current and promising areas of microbiology.

## CHARACTERISTICS OF THE CANDIDATE'S ACTIVITY

Chief Ass. Prof. Ivanka Boyadzhieva is a co-author of 31 scientific papers, of which 27 are scientific articles, 3 are book chapters, and 1 patent. For the competition, she presents 20 scientific articles, 3 chapters of a book, and 1 patent. Twenty articles have been published in journals with IF, the total IF is 42.694, with *h*-index 10 (Scopus).

### Compliance with the requirements of the LDASRB

Group	Content	Requirement	Indicators of Senior Assistant
of indicators		for Associate	Professor Dr. Ivanka Boyadzhieva
		Professor	
A	Indicator 1	50	<b>50</b> p
В	Indicator 2	-	-
С	Indicator 3 и 4	100	100 p

			5 scientific articles (5 in journals with IF and quartile Q2)
D	Sum of indicators	220	340 p
	from 5 до 9		-
	D7		270 p
			15 scientific articles (14 in
			journals with IF; of which 3 with
			Q1, 6 with Q2, 5 with Q3) and 2
			book chapters
	D8		45 p - 3 book chapters
	D9		25 p - 1 patent
Е	Sum of indicators	60	<b>326</b> p - 163 citations (SCOPUS)
	from 10 to 12		

#### Compliance with additional requirements of the SAIM-BAS

The candidate is a co-author of 23 articles after receiving the NOS "doctor" (required 20), and in 6 articles is the first author or corresponding author. The total number of citations is 218 instead of the required 100.

The certificate of fulfillment of the minimum requirements for the academic position "Associate Professor" shows that the candidate scored **816 points**, which covers and exceeds the required 430 points. In addition, Dr. Boyadzhieva also exceeds the additional requirements of the SAIM-BAS.

#### RESEARCH ACTIVITY

The scientific papers presented by Dr. Ivanka Boyadzhieva correspond to the theme of this competition and demonstrate her activity in a current aspect of microbiology, namely, extremophilic microorganisms - biodiversity, biotechnological potential, and application. Four scientific directions are outlined, in which important scientific and applied contributions have been formulated.

#### Biodiversity of microbial communities in extreme niches

Dr. Boyadzhieva is a co-author of scientific papers on the distribution of bacteria and archaea in extreme conditions of existence. In a number of cases, these are the first data on microbial diversity in such habitats in Bulgaria, such as the largest Salt Lake in the Pomorie salt pans and the Mirovo rock salt deposit. Classical and molecular biological methods were used to prove genera and species, some of which are new to microbiological science. I would like to emphasize the relevance of the conducted research and its applied significance.

- For the first time, data on bacterial and aerchial diversity in the P18 crystallizer basin of the Pomorie salt pans and the Mirovo rock salt deposit have been presented.
- A new ratio in the diversity of bacteria and archaea in hypersaline waters has been proven.
- ➤ New representatives of rare and unique genera have been identified, presenting the Bulgarian extremophile microflora to the international scientific community.

## Biosynthetic potential of extremophiles from Bulgarian extreme niches

The unique structural and functional strategies of microorganisms for adaptation to extreme conditions are of growing scientific interest. The importance of extremophiles lies in the possibility of their use in bioprocesses with biotechnological applications, as well as their role as a major source of numerous biomolecules with high added value. In this scientific niche, Dr. Boyadzhieva

presents current and significant developments regarding the synthesis of thermostable and halophilic enzymes from extremophilic bacteria and archaea isolated from Bulgarian habitats.

- ➤ The ability of extremophilic bacteria to synthesize enzymes that degrade 18 different substrates has been established, with the highest frequency of polygalacturonase (91%), catalase and phytase (54%), and lipase (36%) activities.
- For the first time, producers of xanthine lyase, gellan lyase, arabinase, and phytase from the group of halophilic bacteria have been identified.
- ➤ A halophilic microbial community was isolated from a plastic-contaminated area in Burgas Lake, showing the ability to degrade polycaprolactone (PCL), and to a lesser extent, polystyrene (PS) and polypropylene (PP).
- A new thermophilic strain (UG-5B) of the genus Bacillus was identified, which synthesizes the thermophilic enzyme nitrilase, inducible in the presence of benzonitrile.
- $\triangleright$  A strain of the species Chromohalobacter canadensis, a producer of exopolysaccharides, was established. The synthesis of a polymer containing  $\gamma$ -PGA (polyglutamic acid) by a halophilic bacterium was reported for the first time.

## Microbial enzymes and exopolysaccharides of biotechnological importance

This area includes research of high scientific and applied importance. Methods for isolation, purification, and characterization of enzymes and EPZ from extremophilic bacteria have been developed, which can contribute to the development of white and green biotechnologies.

- Extremophilic producers of pectinase (Virgibacillus salaries, Anoxybacillus gonensis), phytase (Cobetia marina 439), nitrilase (Bacillus sp. UG-5B), cyclodextrin-glucanotransferase (CGTase, Bacillus pseudalcaliphilus), and lipase (Brevibacillus thermoruber 7) have been proven.
  - ➤ The following enzymes have been purified and characterized:
    - ✓ Halophilic phytase with application in the feed industry.
    - ✓ Haloalkalophilic polygalacturonase with prospects for treatment of wastewater from the textile, tissue, and paper industries.
    - ✓ Alkaliphilic thermostable pectinase with potential for application in the textile and paper industry, food industry, and processes related to thermal treatment.
    - ✓ Thermostable nitrilase with broad substrate specificity and application in the detoxification of industrial wastewater at high temperature.
    - ✓ Thermostable CGTase, suitable for industrial processes at high temperature and alkaline pH in the pharmaceutical and food industry.
- ➤ A purified EPZ preparation was obtained with significant potential for use in medicine, pharmacy, cosmetics, and the food industry.

## Participation in joint research with teams of scientists working in other scientific fields

The candidate presents multidisciplinary developments in which she participates with her qualification as a microbiologist.

- ➤ It has been proven that cells of Bacillus sp. UG-5B can be effectively encapsulated in sol-gel hybrid matrices composed of tetraethoxysilane and various organic components (such as polyethylene glycol, glycerol, etc.). The resulting matrix is suitable for immobilization of microbial cells with long-term viability and high enzymatic activity.
- ➤ Six types, 12 classes, 21 orders, 43 families, and 106 genera of bacteria were identified in samples from five species of lizards distributed in Western Bulgaria. Species differences in their genetic profiles were proven.

➤ The applicability of bacterial nanocellulose (BNC) and hybrid membranes of BNC impregnated with poly(benzimidazole) (PBI) as environmentally friendly and efficient separators for supercapacitor cells has been demonstrated.

## PARTICIPATION IN PROJECTS

The materials for the competition include evidence of Dr. I. Boyadzhieva's participation in 9 research projects: 3 national, with the National Research Fund at the Ministry of Education and Science; 6 international, under the bilateral cooperation program of the Bulgarian Academy of Sciences (BRAS), with Italy, Turkey, Russia, and Romania, respectively. The candidate is a participant in 8 and is the leader of one.

# CRITICAL REMARKS AND RECOMMENDATIONS

I have no critical remarks on the submitted materials. Given Dr. Boyadzhieva's qualifications in current scientific areas and her skills as a researcher, I recommend that she participate more actively in the development of projects as a leader.

# **CONCLUSION**

The documents and materials presented by Senior Asst. Prof. Dr. Ivanka Petrova Boyadzhieva meets the requirements of the LDASRB, the Regulations for its Implementation, and the relevant Regulations of SAIM-BAS. The candidate has enough scientific papers for the competition, published after the materials used in the defense of the education and scientific degree "doctor". The achieved results in scientific research fully comply with the minimum national requirements and the additional requirements of the SAIM-BAS, adopted in connection with the implementation of the LDASRB.

The scientific papers presented by Dr. Ivanka Boyadzhieva contain original scientific and applied contributions and have received recognition from our and the international scientific community. They are a criterion for extensive professional experience, high qualification, and competence in a current and promising field of microbiology, such as the science of extremophiles and their biotechnological potential. Because of all the above, I confidently give my positive assessment and recommend to the esteemed scientific jury to prepare a report-proposal to the Scientific Council of the Stephan Angeloff Institute of Microbiology at the Bulgarian Academy of Sciences for the election of Chief Assistant Professor Dr. IVANKA PETROVA BOYADZHIEVA to the academic position of "ASSOCIATE PROFESSOR" at the SAIM-BAS in the field of higher education 4. Natural Sciences, Mathematics and Informatics, professional field 4.3. Biological Sciences, Scientific specialty: Microbiology.

Signature:
/Assoc. Prof. Radoslav Abrashev, PhD/