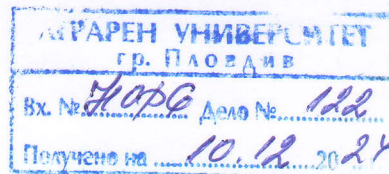


REVIEW



regarding competition for occupying the academic position of “Associate Professor” in the field of higher education: 6. Agrarian Sciences and Veterinary Medicine, professional area: 6.2 Plant protection, scientific speciality: Plant Protection /Microbiology/

CANDIDATE: *Chief Assistant Professor Mariana Krasimirova Petkova, PhD, Microbiology and Ecological Biotechnologies Department, Agricultural University - Plovdiv*

REVIEWER: *Prof. Petar Nikolov Chavdarov, PhD, Institute for Plant Genetic Resources – Sadovo, the field of higher education: 6. Agrarian Sciences and Veterinary Medicine, professional area: 6.2 Plant Protection, scientific speciality: Plant Protection /Phytopathology/, assigned a member of the scientific jury according to Order № ПД 16-1186/ 22.10.2024 of the Rector of the Agricultural University – Plovdiv*

1. Common data regarding the candidate’s career and thematic development

Mariana Krasimirova Petkova was born on November 11, 1971, in Plovdiv. She graduated from the Agricultural University of Plovdiv in 1997 after a five-year course of study acquiring a master's degree in Agroengineering and Plant Protection. She obtained the educational and scientific degree of "Doctor" after four years of study (1999 - 2002) in the Department of "Biochemistry of Pesticides" at Kobe University, Japan. She has successfully defended a dissertation entitled: Structure and Function of New Types of Cytochromes P450 from Higher Plants. On December 19, 2007, with protocol No. 36, the Higher Attestation Commission confirmed the educational and scientific degree of "Doctor" awarded to her in Japan in the scientific speciality 01.06.04 Molecular Biology.

From 2003 to 2006, she worked as a postdoctoral student at Osaka University, Japan, in the Department of Protein Biochemistry. There, she participated in a project entitled Cloning and Functional Analysis of Hacf1 and Hsnf2h Involved in Chromatin Remodeling. Project activities included cloning of both hACF1 and hSNF2h genes and expression in Baculovirus, protein purification, and functional analysis. Since 2010 the candidate has worked as an assistant professor, and since 2011 - as a chief assistant professor, at the Department of Genetics and Selection of the Agricultural University of Plovdiv, where she has led classes in *Genetics* and *Molecular Genetics* with third-year full-time students majoring in Plant Biotechnologies, as well as classes in *Recombinant DNA Technologies* with fourth-year students majoring in Plant Biotechnologies.

Since 2014, Mariana Petkova has been working as a chief assistant professor in the Department of Microbiology and Environmental Biotechnologies at the Agricultural University of Plovdiv. She leads classes in Microbiology. She developed and launched the discipline entitled Molecular Methods for Analysis in Plant Protection for Integrated Plant Protection Master’s degree program. She has improved her qualifications with participation in training courses and seminars related to the application of modern analytical approaches for

ensuring food quality and safety, beverages and forages, HPLC, and food traceability technologies based on DNA analysis and metagenomic analyses.

2. General description of the submitted materials.

The candidate has submitted all necessary references and information according to the Act on the Development of the Academic Staff in the Republic of Bulgaria (ADASRB) and the Regulations for its Implementation of the Agricultural University of Plovdiv.

▶ *Scientific publications under the nomenclature speciality – 43, grouped as follows:*

▶ *Publications related to the dissertation work – 2, which are not subject to the present review;*

▶ *Publications with impact factor – 20;*

▶ *Publications in refereed and reviewed scientific journals – 14;*

▶ *Publications in conference collections – 7.*

The personal participation of Chief Asst. Prof. Mariana Petkova in the mentioned 41 works is illustrated by the fact that one article is independent, in 18 - she is the first author, in 9 - she is the second author, and in the other 13 - she is the third and subsequent author.

Chief Asst. Prof. Mariana Petkova participates in the present competition with 20 scientific papers and reports published in journals with an impact factor and impact rank distributed as follows by quartiles: **Q1 - 3, Q2 - 7, Q3 - 3, and Q4 - 7**, with a total **IF = 34.824**. All this is evidence of the candidate's high scientific contribution in the conducted research studies.

The data presented in Table 1 show that the candidate's scientific production fully meets the minimum national requirements for holding the academic position of "Associate Professor". The increase in the total number of points by 232.3 is very impressive, which is due to the higher number of points from indicators B, Γ and Д.

Table №1 Number of points by indicators. The data presented in Table 1 show that the candidate's scientific production fully meets the minimum national requirements for holding the academic position of "Associate Professor". The increase in the total number of points by 232.3 is very impressive, which is due to the higher number of points from indicators B, Γ and Д.

Table №1 Number of points by indicators.

Group	Indicator	Required number of points	Number of points
A	1. A dissertation work for acquiring the educational and scientific degree of "Doctor"	50	50
B	4. A habitation thesis – scientific publications (no less than 10) in journals refereed and indexed in world databases with scientific information	100	119.5
Γ	7. Articles and reports published in scientific journals refereed and indexed in world databases with scientific information	200	237
	8. Articles and reports published in non-refereed journals with scientific review or in editors'	The sum of indicators from Γ5 to Γ12	19.8

	collective volumes		
Д	13. Citations or reviews in scientific editions refereed and indexed in world databases with scientific information or in monographs and collective volumes	50 The sum of indicators from 13 to 15	86
	14. Citations in monographs and collective volumes with scientific review		33
	15. Citations or reviews in non-refereed journals with scientific review		2
Е	18. Participation in a national research or educational project	100 The sum of indicators from E16 to the end	45
	21. Leadership of an international research or educational project		40
TOTAL		400	632.3

3. Main directions in the candidate`s research work. Skills demonstrated in leading research studies (projects, external financing, etc.)

All research studies have been conducted in three main directions:

1. *Molecular identification, biochemical characterization and in vitro study of the antimicrobial activity of beneficial microorganisms (lactic acid bacteria, yeasts, entomopathogenic fungi of the genus Beauveria) against phytopathogens and pests.*
2. *Study on the influence of the examined beneficial microorganisms on plant growth and development with the aim of their application as biofertilizers in agriculture.*
3. *Application of next-generation sequencing in studying the microbiome of different soils and compost.*

The research studies of Chief Asst. Prof. Mariana Petkova from the first and second groups are based on the identification and characterization of 43 isolates of lactic acid bacteria from fermented plant-based food products. Molecular identification to the species level was performed by amplification of the 16S rRNA gene and subsequent sequencing of 16S rDNA. Screening for exopolysaccharide synthesis was performed with all 43 strains (**В7, Г8 and Г13**).

Chief Asst. Prof. Petkova proves that yeasts exhibit beneficial properties. The research studies in this area are related to the examination of endophytic strains, physiological-chemical, genetic and cultural characteristics to prove PGP-activity, with a view to their application in organic agriculture (**Публикации В5, В6, Г7**).

In recent years, some of the candidate`s studies have been related to the assessment of the potential of entomopathogenic fungi for both phytopathogenic fungi and their insecticidal activity. Endophytic strains have a growth-stimulating effect on colonized plants and outperform non-inoculated controls (**Publications В4, В6 and Г7**). Among the mechanisms involved in growth stimulation is the production of phytohormones (auxins - indole-3-acetic acid) associated with the elongation, division and differentiation of plant cells and the regulation of plant defence responses. In a study conducted in 2020, the candidate found that

plants developed from tobacco seeds inoculated with *B. bassiana* have enhanced growth with an increased rate of photosynthesis, high chlorophyll content and greater density of stomata and trichomes (**Publications B4, B6**). Colonized plants also showed significantly increased tolerance/resistance against bacterial and fungal pathogens. Colonization of potato tubers with *B. bassiana* also showed higher resistance to Colorado potato beetle compared to untreated controls (*Leptinotarsa decemlineata* Say) (**Publications B4 and B5**).

The third group of studies includes metagenomic analyses of microbial communities in microbial processes, which is essential for agriculture. The results of these studies make it possible to understand in detail the microbial interactions (**Publications Г12 and Г20**).

As a participant in a project related to the application of metagenomic and biotechnological approaches for the sustainable use and conservation of some wild species of the *Fabaceae* family in the Strandzha Nature Park region, Chief Assistant Professor Mariana Petkova is studying the bacterial communities in the rhizosphere of chickpea species endemic to this region (*Cicer montbretii* Jaub & Spach) (**Publications Г12 and Г20**). The study aims to establish the relationship between the species diversity of microorganisms and the geographical origin of leguminous plants.

From 2010 to 2013 the candidate was the operational manager of project 10-11 at the National Institute of Science and Technology on the topic: "Molecular-genetic approaches for improving the quality of rapeseed oil for food and industrial needs". He won a scholarship from the Norman Borlaug Foundation at North Dakota State University, Institute of Sunflower Genetics and Selection and Plant Biology, where she worked on the research study entitled: "Genetics and Biochemistry of Rapeseed".

During the period 2014-2024, she was the manager of 4 internal projects funded by the Agricultural University - Plovdiv and one international project under the INTERREG Balkans-Mediterranean Transnational Cooperation Program, 2014-2020. She participated in 3 internal, 2 international and 2 national projects.

4. Evaluation of the candidate`s teaching preparation and activities. Her role in the training of young specialists.

Chief Asst. Prof. Mariana Petkova leads General Microbiology classes to second-year full-time and part-time students majoring in Plant Protection, Environmental Protection, Selection and Seed Production, Zoo-engineering, General Agronomy, Agronomy of Tropics and Subtropics, Ornamental Horticulture, etc.; Ecology of Microorganisms classes to second-year full-time and part-time students majoring in Plant Protection and Ecology and Environmental Protection; Microbial Preparations in Fruit Growing classes for students in the Master's degree program; Molecular Biology lectures and classes in the Master's degree program Integrated Plant Protection; Introduction to Agrarian Education lectures and classes to first-year students in full-time and part-time study majoring in Plant Protection and Ecology and Environmental Protection. During the period 2014-2024, a total of 4348.3 academic hours were completed. On an annual basis, the volume of academic workload averages 434.83 hours, which meets the regulatory requirements for the number of hours at full load.

A syllabus has been developed for the discipline "Molecular Methods for Analysis in Plant Protection" for the Integrated Plant Protection Master's course at the Faculty of Plant Protection and Agroecology. The syllabus includes topics such as molecular genetics, molecular biotechnology, PCR-dependent techniques for detection of microorganisms, bioinformatics analysis and others. Laboratory classes introduce students to modern methods for isolation and analysis of DNA, RNA and proteins, PCR, SDS-PAGE and Western blot, enzyme analysis. Students develop skills in revealing the essence of the molecular mechanisms that control life processes, which in turn leads to an understanding of the methods used for the identification and molecular characterization of microorganisms.

Students have been included in the research activities on projects aiming at their higher quality training. For the same period, the candidate supervised 7 graduates in bachelor's and master's courses, who have successfully defended their diploma theses. Since 2016 Chief Asst. Prof. Mariana Petkova was the coordinator of the Erasmus+ program for the selection of students. To encourage students to participate in the research activities of the Agricultural University, Chief Asst. Prof. Mariana Petkova was repeatedly the course supervisor of bachelor students in Plant Protection and Agroecology.

5. Significance of the obtained results proved with citations, publications in renowned journals, awards, membership in international and national scientific bodies, etc.

The main study results obtained by Chief Asst. Prof. Mariana Petkova has been published in renowned international journals, refereed and indexed in Scopus and Web of Science. She has presented seven reports at various national and international conferences. She was a lecturer at AGBIOL, 2023 with a report: *Endophytic microorganisms for plant growth promotion and biotechnological applications*, at an international conference on agriculture, biological and natural sciences, Edirne, Turkey. The significance of the results was proven by 121 citations: 86 in journals with an impact factor, 33 - in peer-reviewed journals without an impact factor, and 2 cited in doctoral dissertations.

She has been a member of the Union of Scientists in Bulgaria since 2011. As the supervisor of the graduate Konstantina Tsaruhova, who presented a report on the topic "Solution of phosphorus and nitrogen fixation by microorganisms isolated from the rhizosphere of *Cicer montbretii*", she received first prize from the youth forum: "Science, Technology, Innovation, Business", spring 2023.

6. Scientific and practical significance of the contributions. To what extent the candidate has a clear profile of her research work?

I fully accept the presented comprehensive report of the contributions from the research studies. In my opinion, some of the main contributions can be grouped as follows:

I. Scientific contributions of original nature:

► The role of amylolytic lactic acid bacteria in suppressing microbial spoilage of spontaneously fermented doughs has been established (**Publications B2, B7, Г7-5 and Г8**).

► Strains of *L. plantarum* and *L. brevis* have been isolated and identified, capable of suppressing in vitro the development of *Botrytis cinerea*, *Pseudomonas syringae* pv. *syringae* on grapes and *Rhizopus stolonifer* on strawberries (**Publications B7 and Г7-13**).

► The presence of one or more genes encoding the synthesis of bacteriocins, plantaricins and brevacins in *L. plantarum* and *L. brevis* has been proven (**Publications B7 and Г 7-13**).

► The role of endophytic yeasts *Saccharomyces cerevisiae* *Zygosaccharomyces bailii* and *Saccharomyces kudriavzevii* on the growth and development of plants from the *Solanaceae* family has been studied (**Publications B6 and B14**).

► The antimicrobial activity of yeast strains *Pichia fermentans* YP6 and *Saccharomyces cerevisiae* YBS14 against plant-pathogenic fungi, as well as endophytic colonization of plants from the *Solanaceae* family by soil and foliar treatment was determined (**Publication Г 7-14**).

► The effect of microbial endophytes on photosynthesis, stomatal conductance and transpiration intensity in tobacco was studied (**Publication B6**).

► The potential of selected strains of monophytic beneficial microorganisms to influence plant growth and development was established (**Publications B4, B5, B7, B8, Г7-7, Г7-13**).

► The antimicrobial effect of endophytic yeasts against phytopathogens (*Fusarium solani*, *Alternaria solani* and *Rhizoctonia solani*) on plants from the *Solanaceae* family was established (**Publications B6 and Г14**).

► The potential of the entomopathogenic fungi *Beauveria bassiana* and *Beauveria brongniartii* to endophytically colonize various tissues of tobacco was established (**Publications B6 and Г7-7**).

► The efficacy of *Beauveria bassiana* strain 339 against adults and larvae of the Colorado potato beetle (*Leptinotarsa decemlineata* Say) in laboratory conditions was established (**Publication B5**).

► The efficacy of *Beauveria bassiana* 214, 644 and 733 against *Tettigonia viridissima* L. was established (**Publication Г7-23**).

► For the first time in Bulgaria, the microbiome of soils from the locations of wild legume species (*Cicer montbretti* and *Lupinus albus*) was established (**Publications Г7-16, Г7-20 and Г8-7**).

► For the first time in Bulgaria, prokaryotic and eukaryotic microbiomes were established in the different stages of composting of bio-waste on sites with different characteristics (**Publications Г7-10 and Г7-11**).

► The first report of powdery mildew caused by *Erysiphe cruciferarum* on camellia (*Camelina sativa* L. Crantz) in Bulgaria has been announced (**Publications Г7-22**).

II. Scientific contributions of applied and methodological nature:

► A collection of strains of monophytic beneficial microorganisms (lactic acid bacteria, fungi of the genus *Beauveria*, and yeasts) has been created for future applications in plant breeding.

► A methodology for REAL-TIME PCR screening of amylolytic and probiotic strains of lactic acid bacteria has been developed (Publication Г7-5)

► It has been proven that the expiry date of fresh table grapes can be extended by 7-10 days, that of strawberries - by 3-5 days under normal conditions. The ability of lactic acid

bacteria to suppress the development of the phytopathogenic fungus *B. cinerea* is of great importance for the production of healthier food raw materials (Publications Г13, Г7-12).

► A collection of unique amylolytic strains isolated from Bulgarian spontaneously fermented yeasts has been created for future applications in the production of functional foods and food supplements (**Publication B2, B7 and Г8**); A utility model has been created, for which a protected document has been issued with a unique number BG 3805 U1/ 11.05.2020.

► The insecticidal effect of *B. bassiana* strains 214, 644 and 733 against *T. viridissima* was established. Larval survival was monitored at 5-day intervals until the 15th day after treatment with a suspension of spores of the tested *Beauveria* strains. The highest virulence of the tested fungi was shown by strain 644, followed by strains 214 and 733 (**Publication Г7-23**).

The main results and conclusions from the conducted studies have important applied aspects that will increase awareness of potential applications of endophytic microorganisms in biological control systems in organic agriculture.

7. Critical notes and recommendations.

I have no critical notes or questions about the submitted documents and the candidate's overall research production.

8. Personal impressions and reviewer's opinion.

I have known **Chief Asst. Prof. Mariana Petkova** since our common participation in various scientific conferences and seminars. I have also shaped my opinion after the mutual participation in research studies and projects with the candidate and other colleagues from the Institute of Plant Genetic Resources. I consider that the candidate possesses high professional background in the field of molecular biology and microbiology.

9. Conclusion.

The documents regarding the present competition submitted for review show that the research, teaching, applied and publication activities of **Chief Asst. Prof. Mariana Petkova** complies with the Act on the Development of the Academic Staff in the Republic of Bulgaria and the Regulations for its implementation in the Agricultural University - Plovdiv.

All this gives me reason to evaluate **POSITIVELY** the candidate's overall work.

I would like to propose to the honourable Scientific Jury to also vote positively, and the Faculty Council of the Faculty of Plant Protection and Agroecology at the Agricultural University - Plovdiv to confer the academic position of "**Associate Professor**" on **Chief Asst. Prof. Mariana Petkova, PhD**, in the field of higher education: 6. Agrarian Sciences and Veterinary Medicine; professional area: 6.2 Plant Protection; scientific specialty: "Plant Protection (for the needs of the Department of Microbiology)".

3th December 2024
The town of Sadovo

REVIEWER:
/Prof. Petar Chavdarov, PhD/