

REVIEW

by Prof. Velizar Kostadinov Gochev, PhD,

Head of Department “Biochemistry and Microbiology”,

Faculty of Biology, Paisii Hilendarski University of Plovdiv

of the materials submitted for participation in a competition

for the appointment to the academic position “**Associated Professor**” at the Paisii Hilendarski

University of Plovdiv in: higher education area 5. Technical sciences

professional field 5.11. Biotechnologies (Ecological and agrobiotechnologies)

1. General description of the materials provided in the competition

By order No. RD - 21-335 of February 15, 2023, of the Rector of the University of Plovdiv I have been appointed as a member of the scientific jury in the competition for the appointment to the academic position “Associated Professor” at the Paisii Hilendarski University of Plovdiv in: higher education area 5. Technical sciences, professional field 5.11. Biotechnologies (Ecological and agrobiotechnologies), announced in the State Gazette No. 92 of November 18, 2022, for the needs of the Department of Biochemistry and Microbiology at the Faculty of Biology, University of Plovdiv. The only candidate who has applied for participation in the announced competition is Senior Asst. Prof. Ivan Zlatkov Iliev, PhD. The set of materials, provided by the candidate in printed and electronic form complies with the Regulations on Academic Staff Development at the University of Plovdiv (RASDUP), and contains the following documents:

- application form to the Rector of the University of Plovdiv for admittance to the competition;
- curriculum vitae, in the European format;
- higher education diploma for acquiring the educational and qualification degree “Master of Science”;
- diploma for the educational and scientific degree “Doctor” (PhD);
- lists of scientific publications;
- reference to the implementation of the national minimum requirements according to ASDARB and the specific requirements of the University of Plovdiv and FB according to RASDUP;
- habilitation thesis - monograph, copies of scientific publications, citations, handbooks, textbooks, ECTS courses, etc., certifying the implementation of the national minimum and

specific requirements of the FB for the appointment of the academic position "Associate Professor";

- author's reference for the contributions of the scientific publications;
- declaration of originality and authenticity;
- documents certifying the educational activity, scientific and work experience.

The applicant, Senior Asst. Prof. Ivan Zlatkov Iliev, PhD, has enclosed one habilitation thesis, 32 scientific works, one textbook, one handbook, and two copyright certificates for the development and practical implementation of technologies, as well as 7 ECTS courses for the educational and qualification degrees "Bachelor" and "Master of Science". The presented materials do not duplicate any material related to the dissertation for the acquisition of the PhD, and the procedure for the appointment to the Senior Assistant Professor position, due to which all of the materials submitted are accepted for review.

The content and the way of presentation of the submitted materials give me grounds to define categorically the current procedure as lawful.

2. Short biographical information about the candidate

In 2001 Ivan Zlatkov Iliev acquired a Master's degree in Biology - specializing in Plant Biotechnology from the Faculty of Biology of the University of Plovdiv. From 2006 to 2011 he worked as an Assistant Professor at the Institute of Fisheries and Aquaculture (IFA) of the Agricultural Academy. In 2011 he joined the University of Plovdiv as an Assistant Professor in the Department of Biochemistry and Microbiology. In 2015, after a successful thesis defense on "Microbiological and hydrobiological status in the area of net cage aquaculture farms in the Kardzhali reservoir, Bulgaria.", he was awarded the educational and scientific degree "Doctor" in the doctoral programme of Microbiology. In the same year, he took the academic position of "Senior Assistant Professor". Since 2015 till now, Senior Asst. Prof. PhD Ivan Iliev gives lectures and exercises in the disciplines of *Biotechnology of renewable energy sources* and *Microbial energy sources* and exercises in *Microbiology, Microbial metabolism, Microbial ecology, Bioremediation, Microbiological diagnostics*, and practices in *Microbiology and Microbiological diagnostics*.

The professional biography of the candidate is entirely within the scope of the announced competition and it is closely integrated with the primary division, which declared the need for the current competition.

3. General characteristics of the candidate's activity

Assessment of the candidate's research activity

Group of indicators A: Indicator 1 Dissertation for the acquisition of the educational and scientific degree "Doctor" – requirement of ASDARB – 50 pts., implementation by the candidate – 50 pts.

Group of indicators C (The sum of indicators 3 or 4):

Indicator 3 Habilitation thesis-monography or Indicator 4 Scientific publications in scientific editions, which are referenced and indexed in the world databases with scientific information Scopus and Web of Science – requirement of ASDARB – 100 pts., implementation by the candidate – 100 pts.

The monographic work of Senior Asst. Prof. Ivan Iliev, PhD, presents a comprehensive perspective on the solution of a complex and significant problem not only for our country, which is the sustainable development of the aquaculture sector in complex, heavily modified water bodies (HMWB). Up to date, no such study for the country has been presented in the specialized literature. The topic is relevant, considering the place of the sector in conditions of highly limited water resources, in the absence of a clearly dominant type of water usage, where the interests of a number of sectors (tourism, irrigation, electricity generation, etc.) intersect. The paper presents a validated methodology for determining the pressures from net-cage aquaculture on water bodies at a point in time when the government policy is to achieve a good ecological status of surface waters in Bulgaria, according to the regulatory requirements of the Water Framework Directive (2000/60/EU) (WFD). In the spirit of all that has been said, I categorically define the topic of the monograph as particularly up-to-date.

The monographic work has a total volume of 178 standard A4 pages. At the outset, basic provisions setting out the general principles for the determination of the HMWB ecological potential and the related legal requirements and regulations in Bulgaria are described. The existing classification systems for determining ecological potential (EP) in Bulgaria and the relative role of biological, physicochemical, and hydromorphological quality elements in the process are discussed.

In the next part, the EP of the nine HMWBs within the scope of the habilitation thesis is assessed based on hydromorphological and physicochemical quality elements. In support of his work, the author presents a significant set of own empirical data, which in the course of the presentation, is supported by an in-depth theoretical analysis and information collected from the available databases from the control/operational monitoring of the National Environmental Monitoring System (NEMS) for almost ten years period.

The interdisciplinary approach chosen is impressive, where the ecological potential thus identified is used as a starting point for testing the reliability of bacterioplankton as an additional biological quality element (BQE). In the last decade, with the development of molecular methods for taxonomic analysis, increasing attention has been paid to the key role of the microbiome in self-purification processes and the functioning of biogeochemical cycles in aquatic ecosystems. The interest is driven by the fact that microorganisms, like phytoplankton, are among the first communities to respond to changes in environmental conditions, making them a suitable candidate as BQE.

The habilitation thesis presents a first systematic analysis of bacterial communities in Bulgarian HMWBs in addition to the four intercalibrated for Bulgaria BQEs, operationally defined in the Water Framework Directive. Two main aspects of bacterial self-purification, the presence of basic sanitary indicator microorganisms (*Escherichia coli*; faecal coliforms; faecal streptococci and total bacterial viable count) at chosen points along the longitudinal axis of the water bodies, were assessed to identify the point and diffuse sources of anthropogenic pressure; analysis of variations in the taxonomic composition and community-level physiological profiles of bacterioplankton using modern microbiological, biochemical and molecular biological approaches.

Considerable attention is paid to the problems associated with the development of net-cage fish farming in complex reservoirs. The technology of fish rearing in net cages, as well as the characteristics of this type of aquaculture, the available permits, and the quantities of produced fish, are described in detail. The author presents a methodology, tested and validated with his leading role, focused on the determination of the capacity of water bodies to assimilate nutrient pollution introduced as a result of the implemented aquaculture technologies. Maximum fish production quotas are set for each of the nine HMWBs and directions are developed to ensure the sustainable development of the sector.

The submitted work "Technology for ecological assessment and planning of sustainable aquaculture in complex and significant reservoirs" is constructed on the basis of own original unpublished results, obtained in the course of a ten-year period of research of heavily modified water bodies by Senior Asst. Prof. Ivan Iliev, PhD, and meets the criteria for a "monograph" specified in the ASDARB.

A significant achievement of the author is the demonstration of the low-level reliability of the existing classification system in the process of convergence of HMWB to a certain "lake" type based solely on hydromorphological and geographical characteristics, and the confirmation of the need for the development of a new one. Another significant contribution is

the implementation of the first comprehensive study in the country on the applicability of microbiological indicators as BQE in the ecological assessment of complex reservoirs. As a result, an adapted classification system for water quality assessment of reservoirs in Bulgaria was developed. At the suggestion of the author, the parameters *Escherichia coli*, faecal coliforms and faecal streptococci are, since 2017, a mandatory BQE for analysis included in the permits for the use of surface waters for the purpose of aquaculture and related activities issued by the Ministry of Environment and Water.

In conclusion, undoubtedly one of the greatest merits of the monograph is the proposed pragmatic approach in the development of a technological scheme for the assessment of "Ecological Capacity" for fish production of HMWBs with a clear construction based on a mass balance model, validated by a complex analysis of hydrological, physicochemical and biological quality elements.

Group of indicators D (the sum of indicators from 5 to 10) – requirement of ASDARB – 200 pts., implementation by the candidate – 270.70 pts.

Indicator 7 Scientific publications in scientific editions, which are referenced and indexed in the world databases with scientific information Scopus and Web of Science.

Regarding this indicator, Senior Asst. Prof. Ivan Iliev, PhD, has submitted in the current competition 25 research articles (D.7_1 – D.7.25), published in *Bulgarian Journal of Agricultural Science, Ecologia Balkanica, Acta Zoologica Bulgarica, ZooNotes, Current Bioactive Compounds, Biotechnology and Biotechnological Equipment, International Journal of Environmental Science and Technology, Land and Journal of Basic Microbiology*. The papers can be grouped into three main research areas: (1) ecological assessment of surface waters based on microbiome analysis (publications D.7_1, D.7_2, D.7_3, D.7_6, D.7_7, D.7_14, D.8_1 and D.8_2); (2) Microbial ecology and assessment of bioremediation potential of soil microbial isolates (publications D.7_5, D.7_8, D.7_9, D.7_10, D.7_12, D.7_5 and D.8_3) and (3) development of technologies for the rearing of economically significant hydrobionts (publications D.7_4, D.7_15, D.7_16, D.7_17, D.7_18, D.7_19, D.7_20, D.7_21, D.7_22, D.7_24 and D.8_7). All three research areas are closely related and complementary, allowing the compilation of a comprehensive ecological assessment of the ecological potential of the HMWBs and the determination of their carrying capacity for the production of economically significant species of hydrobionts.

In the first research area, the following major scientific and applied contributions of an original nature can be highlighted:

➤ For the first time in the country, the applicability of microbiological indicators as quality

elements in the environmental assessment of complex reservoirs in addition to the four intercalibrated in the national BQE classification was demonstrated. A classification system adapted for the country was developed to assess water quality according to the abundance of the sanitary indicators total heterotrophic count (TVC), *Escherichia coli*, fecal coliforms (FC), and fecal streptococci (FS).

- For the first time in the country, a comprehensive taxonomic analysis of bacterioplankton was performed by next-generation sequencing (NGS) of the 16S rRNA gene in eleven complex and economically important dams in Bulgaria (Alexander Stamboliiski, Ogosta, Studen Kladenets, Ivaylovgrad, Koprinka, Zhrebchevo, Dospat, Batak, Tsankov Kamak, Ovcharitsa and Aheloy). The presence of a similar dominant complex was found in all dams, with the differentiation of water bodies resulting only from differences in the relative abundance of taxa.
- For the first time, the term "carrying capacity" was introduced for complex reservoirs in Bulgaria. A methodology for assessment of the "Ecological Capacity" for net-cage aquaculture in the reservoirs was tested and validated, based on a black box mass balance model, essentially a budget of the amount of phosphorus entering and leaving the reservoir.
- For each of the studied complex reservoirs, specific values for the environmental capacity for fish production in net cages were proposed, which were accepted by the Ministry of Environment and Water as the limit for the respective dam when issuing or renewing permits for the use of surface waters for aquaculture and related activities.

In the second research area the following major scientific and applied contributions of an original nature can be highlighted:

- The structure and metabolic profile of the rhizosphere microbiome of lettuce (*Lactuca sativa* L.) grown in unheated greenhouses were analyzed for the first time. Using 16S rRNA NGS and assessment of the community-level physiological profiles, it was found that fertilization strategy has a direct relationship with bacterial taxonomic composition.
- For the first time, the microbiome of protected wetlands along the Maritza River was investigated by 16S rRNA NGS (MiSeq Illumina) sequencing. The results demonstrate that the main determinants of the bacterial community are wetland type and soil type. Sediment communities of constructed areas subjected to periodic drainage are characterized by lower values of biodiversity indices.

In the third research direction the following more important contributions of an original nature can be highlighted:

- A technology for net-cage fish farming of channel catfish (*Ictalurus punctatus* Raf.) was adapted for the conditions in TPP cooling reservoir providing higher annual growth by extending the active feeding period due to maintaining a temperature above 10°C .
- Optimized stocking density of carp (*Cyprinus carpio*) and bighead carp (*Aristichthys nobilis*) in polyculture with herbivorous species in earthen-type ponds was proposed. The best results related to increased survival rate were found for stocking densities of 3:1 carp: bighead carp at the fingerling stage.
- The inclusion of European catfish (*Silurus glanis*) as a biomeliorator in carp farming was found to generate additional yield at the expense of peaceful fish species. It was established that in the presence of weed fish species (*Pseudorasbora parva*) in the polyculture, *S. glanis* exhibits selectivity towards it at the expense of carp. A feeding coefficient for *S. glanis* of 4.3 - 4.7 was achieved when feeding on carp alone.

Group of indicators E (the sum of indicators 12 and 14) – requirement of ASDARB – 50 pts., implementation by the candidate – 1414 pts.

Senior Asst. Prof. Ivan Iliev, PhD, has provided a list of 134 independent positive citations of his works in the systems of *Scopus* and *Web of Science* and 37 citations in non-referenced peer-reviewed journals, which is an excellent testimonial for his results and their impact on scientific communities. According to *Scopus*, the candidate's h-index is 6.

I categorically assess the scientific, applied science, and applied contributions in the research works, submitted for participation in the competition, as original.

Assessment of the educational and pedagogical activity of the candidate

From the submitted reference for the academic work of the candidate it is evident that for the last 6 academic years Senior Asst. Prof. Ivan Iliev, PhD, has been teaching an average of 529 hours, equivalent to exercises in the Bachelor's degree and an average of 116 hours, equivalent to exercises in the Master's degree, which repeatedly exceed the required norm of 360 hours per year. For participation in the competition, the Senior Asst. Prof. Ivan Iliev, PhD, has also submitted 7 ECTS courses for the disciplines Biotechnology of renewable energy sources, Soil microbiology, Water microbiology, Practicum in Microbiology and Practicum in Microbiological Diagnostics for students in "Pharmaceutical Biotechnology" and

"Microbiology and Virology" Bachelor's degree programme and Microbial Energy Sources and Taxonomy of Industrially Significant Microorganisms for the "Industrial Microbiology" and "Microbiology and Microbiological Control" Master's degree programme. All ECTS courses are comprehensive, well-structured in content and present Senior Asst. Prof. Ivan Iliev, PhD, as an accomplished lecturer. The ECTS courses have been successfully applied in the training of students at the BF and are part of the documentation of the BF in the professional fields 5.11 Biotechnology and 4.3 Biological Sciences, which have been successfully accredited by the NEAA.

For participation in the competition, Senior Asst. Prof. Ivan Iliev, PhD, has submitted one textbook - Biotechnological Processes and Facilities and one workbook - Laboratory Exercises in Microbiology. Both the textbook and the workbook are published by the University Publishing House "Paisii Hilendarski" in co-authorship, and are fully integrated with the programme of the specialty "Pharmaceutical Biotechnology" of the PH 5.11 Biotechnology and all specialties of the PH 4.3 Biological Sciences, studying the basic course in microbiology. Senior Asst. Prof. Ivan Iliev, PhD, has been a scientific supervisor of 8 successfully defended graduates, which also proves his ability to work with students.

The acquired teaching experience and the overall educational and pedagogical activity of the Senior Asst. Prof. Ivan Iliev, PhD, is at a level that guarantees quality training of students.

Additional requirements have been introduced for acquiring the academic position of "Associate Professor" in the Faculty of Biology of the Paisii Hilendarski University of Plovdiv, the fulfillment of which by the Senior Asst. Prof. Ivan Iliev, PhD, is as follows:

- Requirement - The candidate must be the author or co-author of at least 2 textbooks or workbooks in the respective field and specialty of the announced competition; implementation by the candidate – one workbook and one textbook.;
- Requirement - supervision of at least 5 successfully defended bachelor or Master thesis; implementation by the candidate - 8 students successfully defended bachelor or Master thesis;
- Requirement - The candidate must provide evidence of at least 5 years of teaching experience; Implementation by the candidate - 16 years, 5 months and 22 days, of which 7 years, 5 months and 26 days as a Senior Asst. Prof.

- Requirement – Participation in at least 2 scientific projects; implementation by the candidate – participation in 22 projects, of which 7 with national funding under the Ministry of Education and Science and Ministry of Environment and Water and 15 - institutional projects under the PU and Agricultural Academy.
- Requirement – organizational and administrative experience for the last 5 years in the faculty; implementation by the candidate – Senior Asst. Prof. Ivan Iliev, PhD, is a member of the committee for preparation of assignments and commissioning public procurement at PU, a participant in the prospective students campaign from 2020 to date, a member of the organizing committee of the student competition "Journey in Biology", a member of the editorial board of the Journal of Bioscience and Biotechnology, administers the BF website.

All the additional requirements of BF for the academic position of "Associate Professor" are fulfilled by Asst. Dr. Ivan Iliev in a categorical manner.

4. Evaluation of the personal contribution of the candidate

I consider that the personal contribution of the Senior Asst. Prof. Ivan Iliev, PhD, to the realization of the publications submitted for the competition is relevant to the level of his competence and covers all stages of the realization of a successful publication, starting from the generation of the idea, its experimental implementation, presentation and statistical processing of the results and formulation of the conclusions.

5. Critical comments and recommendations

I have no significant critical remarks on the content of the material presented in the competition and the way it was formatted. My only recommendation relates to the possibility of a more summarized presentation of the contributions.

6. Personal impressions

I have known Ivan Iliev since he joined the Department and have not only witnessed, but also directly participated in all stages of his career development. I believe that he is very well prepared theoretically in the field of competition, with excellent organizational and technical skills for working with specialized laboratory equipment, statistical software and excellent English. He is very responsible and loyal in his relations with colleagues and students, and he is able to set clear goals to follow purposefully.

CONCLUSION

The documents and materials presented by the Senior Asst. Prof. Ivan Zlatkov Iliev, PhD, comply with the formal and substantive requirements of the RRDA, the Implementing Rules of the ASDARB and the RASDUP. The candidate in the competition has presented a sufficient number of scientific works published after the materials used in the successful PhD thesis defense, in which original scientific and applied contributions in the field of environmental biotechnologies and agrobiotechnologies can be highlighted. The accomplishments, achieved by Senior Asst. Prof. Ivan Iliev, PhD, in teaching and scientific research activities, fully cover and exceed the specific additional requirements of the Faculty of Biology of PU for the academic position "Associate Professor".

After familiarizing with the materials and scientific works presented in the competition, analysis of their significance and original scientific and applied contributions contained in them, I find it reasonably convincing to give my positive assessment and to recommend the Scientific Jury to prepare a report-proposal to the Faculty Council of the Faculty of Biology for the election of the Senior Asst. Prof. Ivan Zlatkov Iliev, PhD, to the academic position of "Associate Professor" at the Faculty of Biology in the field of higher education 5. Technical Sciences, professional field 5.11 Biotechnology (Ecological and Agrobiotechnology).

28.03.2023

Plovdiv

Reviewer:.....

(Prof. Velizat Gochev, PhD)