

OPINION

From: **Prof. Dr. Valentina Todorova**, Institute of Oceanology "Fr. Nansen" - BAS, member of a scientific jury, according to Order No. 58/01.03.2024 of the Director of IO-BAS for conducting a competition for the academic position of "associate professor" in the Field of Higher Education: Code 4. "Natural Sciences, Mathematics and Informatics"; Professional direction: code 4.3. "Biological Sciences"; Scientific specialty: "Ecology and ecosystem protection"; Scientific direction: "Ecology and molecular taxonomy of marine organisms" in the "Marine Biology and Ecology" Department at the Institute of Oceanology, BAS, Varna, announced in State Gazette No. 1/02/01/2024.

1. Education and career development of the candidate

Nina Stoycheva Dzembekova, the only candidate in the competition for the academic position of "associate professor", completed her higher education with a master's degree in ecology and environmental protection at the Technical University - Varna in 2004 with excellent grades. In 2018, she obtained the educational and scientific degree "doctor" in hydrobiology at IO-BAS, where her main career growth took place as an ecologist (2016), assistant (2016-2018) and chief assistant (2019-present). Her qualification in the scientific specialty of the competition is complemented by four trainings in molecular methods in Japan, Italy and Bulgaria. The candidate fully meets the regulatory requirements for the educational and scientific degree "doctor" (indicator A.1 - 50 points) and relevant experience in the scientific field of the competition.

2. Research activity of the candidate

The submitted documentation for the competition is well organized, precise and supplemented with the necessary evidence, creating a positive general impression of a significant volume and high quality of the achieved scientific results.

2.1. Overview of the publications and citations

Nina Dzembekova participated in the competition with a total of 27 scientific publications, of which 22 were in ranked journals and other publications, referenced and indexed in the internationally acknowledged scientific information databases Scopus and/or Web of Science, distributed as follows: Q1 - 7, Q2 - 8, Q3 - 4, Q4 - 2, issues with SJR - 1. The candidate is the first or second author of 8 (33%) of the indexed publications, which distinguishes her as a researcher with a significant personal contribution to the phytoplankton molecular research. Her

participation in larger teams as a third and subsequent author I evaluate completely positively, as it is indicative of her research cooperation in a broader scientific area and successful integration in the national and international scientific community.

After search in the scientific databases Scopus and Web of Science, I confirm that the candidate fulfils and exceeds the minimum national and IO-BAS requirements according to criteria C.4 "habilitation work-publications" - 132 points and D.7 "publications outside the habilitation work" - 272 points.¹

The reference for citations in scientific publications lists 264 citations, respectively, indicator D.11 is estimated at 528 points and exceeds 9-fold the minimum requirements (60 items). The citations visible in the scientific databases Scopus (340 citations, without self-citations) and Web of Science (475 citations, without self-citations) testify convincingly to the significant impact of the published scientific results and prove in an indisputable way that the candidate in the competition meets the regulatory requirements according to this indicator.

2.2. Most important scientific and applied contributions

The main part of Nina Dzembekova's scientific research and results are in the field of phytoplankton biodiversity in the Black Sea, taxonomy and ecology of potentially toxic and blooming species, distribution and ecology of the Black Sea microbiome, characterization of Antarctic diatom communities, including the identification of new species for science, by combining innovative molecular techniques with classical methods.

I would single out the following as the most significant scientific contributions of an original or confirmatory nature:

1. Pioneering molecular studies of the species diversity of the genus *Pseudo-nitzschia* registered a new representative of the genus for the Black Sea - *P. linea* and a new species for the Bulgarian waters - *P. calliantha* (with proven toxigenic potential in the Black Sea) and a new variety form - *P. pungens* var. *aveirensis*. Refinement of taxonomic affiliation at the species level is important given the cryptic biodiversity present in the genus and the inability to distinguish toxic from harmless species using conventional methods.

¹ Publication 6.3.2-2 is in journal with rank Q2 (2021), not Q3 .

2. For the first time in the Black Sea, metabarcoding was applied to study the biodiversity of phytoplankton resting stages in surface sediments, resulting in the identification of 16 potentially toxic species (12 dinoflagellates, 1 diatom, 1 haptophyte and 2 raphidophytes), as more than half of them are reported for the first time in sediment samples from the Black Sea. The study of phytoplankton cysts is important due to their function as an "archive" of biodiversity and as a potential reservoir for bloom phenomena. A relationship was established between the prevalence and abundance of bloom-causing *Scrippsiella acuminata* cysts in the Black Sea sediments and selected environmental variables such as salinity, temperature and nutrients. The geographic distribution of *Scrippsiella* blooms in the Black Sea indicates that the interaction between planktonic and benthic habitats of this dinoflagellate species gives it the advantage to dominate planktonic communities.

3. For the first time, a parallel study of the distribution of toxic/potentially toxic phytoplankton species (microscopic and molecular methods) and phycotoxins was carried out in the Black Sea. 20 potentially toxic species have been identified, some of which (*Dinophysis acuminata*, *Dinophysis acuta*, *Gonyaulax spinifera* and *Karlodinium veneficum*) had a frequency of 95%. The toxins present were determined, some of them (PTX-1, PTX-13, YTX variants) found for the first time in the Black Sea. Positive correlations between the numbers of some microalgae with certain toxins were deduced, suggesting the toxigenicity of the species *Dinophysis acuta*, *Lingulodinium polyedra* and *Protoceratium reticulatum*. The combined study of phytoplankton biodiversity and phycotoxins enables the evaluation of the toxigenic potential of the Black Sea isolates and the formulation of a statistically based hypothesis about the species producing them.

4. The ability of Black Sea isolates of *Synechococcus* from a depth of 750 m to survive, both under conditions rich in oxygen and light, as well as under aphotic and anoxic conditions, has been experimentally proven, a potential also proven by whole-genome sequencing. This finding provides new evidence to support the existing hypothesis that 'deep-sea red fluorescence' is associated with viable populations of picocyanobacteria in deep anoxic ocean zones. Through molecular methods (whole-genome sequencing, metagenomics and qPCR), a heterogeneous distribution of two *Synechococcus* phylotypes, tentatively designated as 'surface' and 'deep-water', has been established. The "surface" phylotype is specifically adapted to surface coastal waters, while the "deep-water" phylotype is able to survive in the aphotic anoxic conditions of the deep layers.

5. Through an innovative approach - shotgun metagenomics, the vertical structure of the pelagic microbiome in the Bulgarian Black Sea was established: in the oxygen zone, photoheterotrophs and cyanobacteria dominate (*Synechococcus*, but *Prochlorococcus* is visibly absent); the chemocline is distinguished by chemolithotrophic metabolism (*Thioglobus*) and facultatively anaerobic microbes; the hydrogen sulphide zone is characterized by a massive dominance of sulphate-reducing bacteria, few (but detectable) marker genes for methanogenesis, and a large number of unclassified genomes with unpredictable ecology. The research adds new, valuable information about the microbiome of a unique ecosystem such as the Black Sea.

6. The knowledge of the biodiversity of the benthic diatom communities in the Antarctic has been enriched by two species new to science - *Halamphora kenderoviana* sp. Nov. and *Halamphora moncheviana*.

I would single out the following developments as the most important applied contributions:

7. Studies of the population-genetic structure of commercially valuable fish species obtain first data (garfish - *Belone belone*) or complement new information (turbot - *Scophthalmus maximus*, mullet - *Mullus barbatus*) about their genetic differentiation and diversity. The low haplotype and nucleotide diversity found in mullet are associated with overexploitation of the population. In addition, indicators of the health status of the mullet such as pollution with heavy metals and microplastic particles, oxidative stress and antioxidant protection were established. The habitat suitability of the turbot was modelled. The obtained results are essential for the protection and effective management of fish stocks in the Bulgarian Black Sea.

8. The detection of multiple antibiotic resistance genes (ARGs) in different areas of the Black Sea, including deep-sea, using qPCR and metagenomic analyses, revealed the Black Sea as a reservoir of multiresistant bacteria, which are considered a new type of contaminants with an increasing threat to human health.

3. Project activity

Although it is not an indicator subject to quantitative assessment in competitions for the academic position of "associate professor", Nina Dzembekova's project activity is an attestation of diverse research activity and successful scientific cooperation with visible results. The candidate participated in the implementation of 19 projects - 6 national and 13 international, one of which

she coordinated on behalf of IO-BAS: " Black Sea Connect ". In implementation of this project, the Strategic Program for Scientific Research and Innovation in the Black Sea was developed.

CONCLUSION: The subject, volume and quality of the publication output, its high citation rate, as well as the significant project activity convince me in an indisputable way that Nina Dzhebekova has a clearly defined research profile in the scientific field of the announced competition: "Ecology and molecular taxonomy of marine organisms". The results achieved according to the mandatory scientometric indicators fully meet and exceed the requirements under Art. 2b, paras 2 and 3, respectively, to the requirements under Art. 2b, para. 5 of the Law for The Development of Academic Staff in The Republic of Bulgaria, the requirements under Art. 1a, paras 1 and 2 of the Rules for Implementation of the Law, the requirements under Art. 2, para. 4.3 of the Rules in the Bulgarian Academy of Sciences, respectively the requirements under Art. 1a, para. 1 of the Rules in the Institute of Oceanology - BAS.

On these grounds, **I express my positive opinion that Nina Dzhebekova should be elected to the position of "associate professor" in the "Marine Biology and Ecology" Department at the Institute of Oceanology, BAS.**

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Varna

Заличен на основание ЗЗЛД

/Prof. Dr. V. Todorova/