

REVIEW

by: Prof. Dr. Mariela Konstantinova Odjakova-Baytocheva, member of the Scientific Jury, appointed by Order No. 110-OB/28.06.2024 of the Director of IMB, BAS, Assoc. Prof. Dr. A. Gospodinov

REGARDING: competition for the academic position of "Assoc. Prof" in the field of higher education 4. Natural sciences, mathematics and informatics, professional direction 4.3. Biological Sciences - Molecular Biology, for the needs of the "Regulation of Gene Activity" section, IMB, BAS - State Gazette, no. 52 of 18.06.2024

General presentation of the procedure and the candidate

In the competition for associate professor in the field of higher education 4. Natural sciences, mathematics and informatics, professional direction 4.3. Biological Sciences - Molecular Biology, announced for the needs of the "Regulation of Gene Activity" section, IMB, BAS, one candidate - chief assistant Dr. Elena Bozhidarova Kruchmarova, IMB-BAS.

The submitted materials for participation in the competition have been prepared in accordance with the requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria (LDASRB), the Regulations for the Implementation and the Regulations for the Terms and Procedures for Acquiring Scientific Degrees and Holding Academic Positions at the IMB, BAS.

Assistant Professor Dr. Kruchmarova completed her higher education at the University of Chemical Technology and Metallurgy, Sofia, where she graduated in 2010 as a master's degree in biotechnological engineering. In 2018, she defended her PhD thesis at IMB, BAS and obtained the educational and scientific degree "PhD".

Dr. Kruchmarova worked at IMB, BAS successively as a chemist (2010-20118); Assistant Professor (2011-2015); chemist (2015-2018) and Ch. assistant from 2018 to now.

She was on two long-term specializations at prestigious foreign institutions - at the Institut de Biologie Structurale (IBS) (Grenoble, France) and at one of the leading biotechnology companies in Europe - Proteros biostructures GmbH (Martinsried, Germany). She also participated in several short-term courses with a molecular-biological focus (PCR, cell culture, protein crystallization and biomolecular interactions).

Dr. Kruchmarova was a consultant in the preparation of two master's theses (one for a student on the "Erasmus" program) at HTMU - Sofia. In 2023, she was an invited speaker at the 47th FEBS Congress, Tours, France. She participated with oral presentation in more than 10 national and international conferences, and with poster presentations in more than 20.

The PhD thesis of Dr. Kruchmarova was awarded by the Union of Scientists in Bulgaria with a diploma from the competition for high scientific achievements, direction "Scientific achievements

in protected doctoral dissertations by scientists under the age of 35". In 2019, she was awarded the 2018 Eureka Award for Achievement in Science, and in 2020 the 2020 Eureka Award for Young Scientists.

Meeting the minimum state requirements for the academic position Associate Professor

Dr. Kruchmarova is a co-author in 17 publications referenced in Scopus/Web of Science. In the current competition, she participated with 14 publications and 2 patents, which were cited 91 times in Web of Science/Scopus, and this number does not include numerous citations in patents and dissertations. The total JCR-IF of all articles is - 47.536.

The scientific output presented and the scientometric data achieved fully correspond, and in many of the indicators, exceed the minimum requirements for awarding the academic position "Associate Professor", defined in the Regulations for the terms and conditions for acquiring scientific degrees and occupying academic positions at IMB, BAS. Data for meeting the requirements by indicators are presented as follows:

Indicators from group A: dissertation work - 50 points;

Indicators from group B: habilitation work - 100 points (4 publications with Q1);

Indicators from group D: publications in journals with IF/IR- 230 points out of required 200/220 points (4 publications with Q1; 1 - Q2; 2 - Q3; 3 publ. with SJR and 2 patents).

Indicators from group D: works cited - 182 points with required 50/60 points.

Indicators from group E: data on 171.9 points achieved are provided. Dr. Kruchmarova participated and participates in 12 national and 2 international scientific projects and led one scientific project financed by the program for supporting young scientists and doctoral students of the BAS (according to the Regulations for the Application of the LDASRB, for the academic position "Associate Professor" points under indicator E are not required).

A total of 733.9 points were achieved with the required 400/430.

Evaluation of scientific and scientific-applied activity

Dr. Kruchmarova's scientific interests and published scientific results are entirely in the field of the announced competition and are concentrated in three main directions:

- Study of the molecular mechanisms of action of the ORF6 and Nsp13 proteins of the SARS-CoV-2 virus in infected cells. Approaches to taming the cytokine storm;

For the first time, a mechanism is proposed by which the interaction of one of the main participants in the export of mRNAs from the nucleus to the cytoplasm ORF6 with RAE1 leads to genome instability. ORF6 inhibitors have been proposed, which is a key step in the development of potential therapeutics for the treatment of COVID-19. Through in silico screening, potential inhibitors, including natural and synthetic substances, of the Nsp13 helicase of SARS-CoV-2 were investigated and a new antiviral mechanism of action of Ritonavir (Norvir) was discovered. The results obtained show that peptide-based inhibitors are a more promising strategy to block Nsp13

and to fight future variants of the virus. The key role of heparan sulfate (HS) and its proteoglycans in all processes involving signal transduction pathways driven by hIFN γ has been revealed. These data provide an opportunity to develop potential biological products to inhibit overexpressed endogenous hIFN γ , which is one of the key factors in the development of cytokine storm. The importance of low-molecular-weight heparin for the prevention and suppression of the cytokine storm resulting from the development of COVID-19 and other viral diseases is shown

- Investigation of factors affecting the biological activity of human gamma-interferon (hIFN γ) and its production as a recombinant protein.

The first model structures of glycosylated homodimers of full-length hIFN γ were obtained and one of the few MD simulations of glycoproteins described so far in the literature was performed. Nucleic acids were found not to be a co-precipitation contaminant, as commonly believed in the literature, but to be a typical component of bacterial inclusion bodies. A strategy for the soluble production of hIFN γ was implemented for the first time, which involved fusing it to an autohydrolyzing protease. As a result of a wide screening study, optimal conditions for the storage of hIFN γ and its mutant derivatives, characterized by instability and a high tendency to aggregation, were established.

- Thermodynamics of interaction of ionic liquids with the transport protein serum albumin

Ibuprofen-based ionic liquids have been shown to exhibit a pharmacokinetic profile close to that of ibuprofen and offer new opportunities to improve drug delivery. Eight salicylic acid-based ionic liquids have been synthesized and characterized in detail, showing potential for increased efficacy in the treatment of chronic skin diseases due to better drug delivery and sustained release. It has been shown that esterification can affect the affinity and thermodynamics of binding of the anti-inflammatory drug naproxen to serum albumin, enabling the development of new drugs and the improvement of existing ones.

Dr. Kruchmarova's important applied contributions include two patents (European and Bulgarian) dedicated to a method for obtaining mutant hIFN γ forms, which are potential drug candidates for the treatment of diseases associated with the overexpression of hIFN γ . Three mutants were obtained that exhibit greatly reduced biological activity (from 2 to 4 orders of magnitude), exhibit good competitiveness with wild-type hIFN γ , and possess a conserved, even stronger affinity for the interferon receptor. They are potential drug candidates for the treatment of diseases associated with hIFN γ overexpression.

Conclusion

The documents and materials presented by Ch. assistant professor Elena Kruchmarova, PhD, for participation in the competition for the academic position of "associate professor", meet all the requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria (LDASRB), the Regulations for its implementation and the Regulations for the conditions and procedures for acquiring scientific degrees and occupying academic positions in IMB, BAS.

Dr. Kruchmarova has presented a sufficient number of scientific papers and convincing evidence for her scientific and scientific-applied work, illustrating the high quality of her overall academic

activity. All this allows me to give a positive assessment and recommend with conviction to the respected members of the Scientific Council of IMB-BAS to vote positively for the election of Ch. assistant professor Elena Bozhidarova Kruchmarova, PhD, to the academic position "Associate Professor"

22.10.2024

Prof. Dr. Mariela Odjakova