REPORT

by Assoc. Prof. Aneliya Stefanova Kostadinova, PhD
Institute of Biophysics and Biomedical Engineering – BAS
Regarding: competition for the academic position of "Associate Professor",
promulgated in the State Gazette, issue 58/18.7.2025.
Professional field: 4.3. Biological Sciences,

Scientific specialty: Biophysics
Member of the scientific jury appointed by order No. 749/17.9.2025
of the Director of IBPhBME-BAS

Only one candidate from the same department applied for the competition for the academic position of "Associate Professor" for the needs of the Department of Electroinduced and Adhesive Properties at IBPhBME-BAS: Senior Assistant Professor Kamelia Todorova Hristova-Panusheva, PhD. The materials for participation in the competition, submitted by the candidate, are presented in detail in accordance with all the requirements of the DASRBA and the Regulations for its implementation at IBPhBME-BAS.

Biographical information about the candidate:

Kamelia Todorova Hristova-Panusheva graduated from the Faculty of Biology of SU "St. Kliment Ohridski" with a degree in Cell Biology and Pathology in 2005. In 2005 she was appointed as an assistant in the Department of Cell Adhesion at the Institute of Biophysics – BAS. In 2006 she won a competition for a full-time doctoral student at the Department of Electroinduced and Adhesive Properties, Institute of Biophysics and Biomedical Engineering – BAS.

From 2010 to 2012 she was an assistant at the Department of Electroinduced and Adhesive Properties, Institute of Biophysics and Biomedical Engineering – BAS. In 2011 she defended her dissertation on the topic "Modulated interaction on osteoblasts with hydroxyapatite materials", with supervisor Prof. Georgi Altankov, DSc, thereby obtaining the educational and scientific degree "doctor" (PhD) in the scientific specialty "Biophysics" at IBpBME-BAS. She has continued her development as a Senior Assistant Professor in the Department of Electroinduced and Adhesive Properties since 2012 to the present. In the period from 2007 to 2009, Kamelia Todorova Hristova-Panusheva completed three doctoral specializations, at the Martin Luther University, Halle, Germany, and at the Institute of Bioengineering of Catalonia, Barcelona, Spain.

Research activity and scientometric data of the candidate:

Senior Assistant Professor Kamelia Todorova Hristova-Panusheva, PhD, has a total of **28** scientific publications and one book chapter. All of them are referenced in Scopus. According to Scopus, **22** of these publications have been cited 171 times (excluding self-citations), with **an h-index of 6**.

For this competition, the candidate has submitted a total of **19** scientific publications and **1** book chapter. Of these, 9 are in Q1, 1 in Q2, 2 in Q3, and 2 in Q4. In three of the submitted publications, Kamelia Hristova-Panusheva is the first author. A reference with 50 citations in Scopus has been submitted for the competition. All this shows the high quality of the presented scientific publications, as well as the personal participation of the candidate therein.

These scientometric indicators fully meet the requirements for occupying the academic position of "Associate Professor" in the Regulations for the implementation of DASRBA at IBPhBME-BAS, cover and exceed the minimum required points for all groups of indicators, as follows: В – 125, Г – 239, Д – 100.

All publications submitted in the competition reflect significant interdisciplinary scientific research in the fields of: biochemistry, chemical engineering, tissue engineering, biophysics,

nanomaterials, etc. They are the result of projects in which the candidate participated and collaborations with national and international scientific groups. According to the submitted documents, Kamelia Hristova-Panusheva is a participant in 3 national projects, one COST project and one international project with China.

Scientific topics and contributions of the publications:

The scientific results and original contributions in the publications of Sen. Assist. Prof. Kamelia Hristova-Panusheva, PhD, included in the competition, are described in an extended habilitation report. They are relevant and interesting from both a fundamental and an applied perspective for modern biomedicine.

The scientific topics can be divided into four main areas:

1. Research in the field of photothermal anticancer therapy, in combination with various nanoparticles such as graphene oxide, pegylated graphene oxide, gold and silver nanoparticles.
2. Establishment of the biological effects and mechanisms of toxicity of nanoparticles in *in vitro* models of cancer and non-cancer cell lines in order to improve the biocompatibility of materials.
3. Study of the parameters of varying laser irradiation for specific combinations of nanoparticles and cancer cells in order to achieve maximum efficacy of nanomaterial-mediated photothermal therapy.
4. Research on cell adhesion and biocompatibility when treating cells with conventional anesthetics used in surgery, biologically characterized new composite materials (HMDS +/- DND and TIN/TIO2) for application in bone tissue engineering, as well as evaluation of the biocompatibility of an ethanol extract of *Haberlea rhodopensis Friv. in vitro* culture in normal skin cells for potential therapeutic application.

The main scientific contributions from these studies are summarized by the candidate into 2 main groups as follows; (1) contributions included in the habilitation work and (2) contributions outside the habilitation work.

1. **The contributions related to the habilitation work** cover the study of the effect of PEGylated graphene oxide nanoparticles and gold nanoparticles in combination with laser irradiation and are divided into two sub-items A and B. (B1-B5)
    - It has been found that functionalization of GO (known for its photothermal conversion efficiency) with PEG improves the physicochemical properties of graphene oxide and reduces its cytotoxicity, making it a promising tool for synergistic cancer therapy.
    - The synergistic effect between PEGylated graphene oxide and continuous near-infrared (NIR) laser irradiation has been established at the cellular and subcellular level, by monitoring mitochondrial dynamics (ROS generation), actin cytoskeleton organization, and nuclear size (gene expression monitoring).
    - The negative influence of GO and GO-PEG on cardiac contractility in frogs has been proven. Causing stronger arrhythmic contractions, they increase the production of ROS in C2C12 myoblasts, the effects of which are reduced after irradiation with NIR. The studied GO/GO-PEG nanoparticles in rat liver significantly stimulate DAO activity, with this effect being enhanced after irradiation with NIR.
    - An adapted therapeutic strategy for liver cancer has been established by irradiating liver cells with NIR in the presence of GO/GO-PEG nanoparticles. Irradiation with NIR at a wavelength of 808 nm caused a slight increase in temperature, enhancing cytotoxicity, with nGO showing higher toxicity by disrupting cell morphology, reducing proliferation, and increasing ROS levels. In contrast, nGO-PEG more effectively suppressed cell motility and demonstrated improved biocompatibility. Gene expression analysis revealed upregulation of apoptosis-related genes in nGO-PEG-

treated cells, indicating mitochondrial damage, while nGO induced autophagy, as evidenced by the increased expression of BECN1. The results point to different therapeutic potentials: nGO as a potent cytotoxic agent inducing autophagy, and nGO-PEG as a more biocompatible nanoparticle promoting apoptosis.

- The synergistic effect of PEGylated graphene oxide and gold nanoparticles in combination with femtosecond laser irradiation on normal kidney cells and hepatocyte cancer cells has been investigated. It has been found that GO-PEG has better photothermal efficiency under Fs irradiation compared to GO, resulting in a larger temperature increase.

- For the first time, the effect of gold nanoparticles after irradiation with a 343 nm Fs laser at low intensity in HepG2 and normal MDCK cells has been studied. A synergistic photothermal effect at low laser intensity has been demonstrated. The combination of ultrasmall AuNPs and low-intensity femtosecond laser irradiation causes a significant increase in the temperature of the cell environment, which causes a strong reduction in the survival rate (up to 40–46%) of HepG2 cells.

2. **The contributions outside the habilitation work** are related to the effects of halothane on lung cells, characterization of new materials for tissue engineering, study of the biocompatibility of two-layer TiN/TiO2 coatings deposited by DC magnetron sputtering on stainless steel. Study of the safety of total ethanol extract of *Haberlea rhodopensis Friv. in vitro* culture (HRT), as well as research related to the study of the effect of a new plant cystatin on the adhesive behaviour of normal and cancer cells. (Г7.2- Г7.14) and (Г8.1)

- It has been found that halothane, from the group of halogenated hydrocarbons, inhibits FAK in A 549 lung cancer cells, by phosphorylation of paxillin, not by proteolytic changes or inhibition of vinculin and paxillin expression.

- Biologically characterized plasma-polymerized coatings and thin films of hexamethyldisiloxane (HMDS) have also been developed to control cell adhesion, growth, function and differentiation by modulating the surface hydrophilicity of plasma-polymerized films of hexamethyldisiloxane (PPHMDS) by treatment with ammonium plasma for different durations. Extracellular matrix proteins adsorbed on all studied PPHMDS films in a conformation that promotes interaction with endothelial cells, regardless of the duration of ammonium plasma treatment. New data on the biocompatibility of PPHMDS thin films prepared by two different technological regimes and modified with ammonium plasma have been obtained.

- A new composite material based on PPHMDS and detonation nanodiamond (DND) has been created and it has been shown that the addition of DND to the layer improves the efficiency of cell adhesion and protein adsorption. It has been found that the way of incorporating DND into the layers in PPHMDS also affects the surface properties of the composite materials and the behaviour of mesenchymal stem (MSC) and MG63 cells.

- The influence of the elasticity and hardness of composite coatings and thin layers of HMDS/PDMS and DND on cell adhesion, growth, function and myoblast cell differentiation has been established. It has been found that an increase in the concentration of DND particles in the PPHMDS or PDMS matrix leads to a clear increase in the elastic modulus of both types of composite layers and to an improvement in the osteogenic differentiation of MSCs.

- The biological effect of extracts of *Haberlea rhodopensis Friv. in vitro* culture (HRT) for therapeutic use in cosmetics has been established. The extract does not damage cells, nor is there any evidence of free radical generation or genotoxicity. Cellular rejuvenation effects

have been established based on tracking the mitochondrial morphology.

- It has been established that plant cysteines inhibit cell adhesion on breast cancer cell lines via an integrin-independent pathway and are a promising candidate for inhibiting cancer metastasis.

Conclusion

The submitted materials for the competition and the scientometric indicators of Sen. Assist. Prof. Kamelia Todorova Hristova-Panusheva, PhD meet all requirements and cover the minimum required points for all groups of indicators from the Regulations for the implementation of the DASRBA at IBPhBME-BAS. I firmly believe that Sen. Assist. Prof. Kamelia Todorova Hristova-Panusheva, PhD, with her competence and scientific production, is a very suitable candidate for the academic position of "Associate Professor" in the Department of Electroinducible and Adhesive Properties at IBPhBME-BAS.

All of the above gives me reason to give my positive assessment with full conviction and to recommend to the Honorable Scientific Jury to prepare a proposal to the Scientific Council of IBPhBME-BAS for the election of Sen. Assist. Prof. Dr. Kamelia Todorova Hristova-Panusheva, PhD to the academic position of "Associate Professor" in professional field 4.3 "Biological Sciences", scientific specialty "Biophysics".

21.11.2025 Signature:

Sofia /Assoc. Prof. Aneliya Kostadinova, PhD/