TO
Chairman of the Scientific Jury
Order No. I-57/23.04.2024
Director of the Institute of Microbiology
"Stefan Angelov" – BAS

Opinion

by Prof. Dr. Maria Mitkova Orbetzova, PhD,
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Head of the Department of Endocrinology and Metabolic Diseases, Medical Faculty,
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External member of the Scientific Jury according to the Order No. I-57/23.04.2024 of the Director of the Institute of Microbiology "Stefan Angelov" - BAS concerning the competition for the academic position of "Associate Professor" in the field of higher education 7. Health and Sport in the professional field 7.1. "Medicine", announced in State Gazette No. 28 of 02.04.2024 for the needs of the Department of Biotechnology, Laboratory of Metabolomics of IMicB - BAS.

1 candidate participated in the competition: Dr. Lilia Vladimirova Mihaylova (Vassileva), MD

I. Documents submitted for the competition:

The applicant has submitted the following documents, which have been accurately prepared and arranged in accordance with the requirements of the competition:

1. CV in European format;
2. Diploma for educational and scientific degree "Doctor"
3. Abstract of the thesis for the educational and scientific degree of Doctor ;
4. Certificate of experience in the specialty;
5. List and copies of publications;
6. List and copies of participation in scientific conferences and supporting material;
7. List of noted citations;
8. Information on participation in research projects ;
9. Brief information about research work with main contributions and views for further research work;
10. Reference on the fulfilment of the minimum requirements according to the Law on the Development of the Academic Staff in the Republic of Bulgaria ;
11. Reference for fulfillment of additional requirements according to the regulations of IMicB-BAS;
12. Summaries in Bulgarian and English of the publications included in this competition;
13. Declaration of originality and reliability of the submitted results;
14. Additional information on scientific activity - list and supporting material;
15. A copy of the State Gazette No 28 of 02.04.2024 with the competition notice;
16. Regulations of the IMicB - BAS on the conditions and procedure for acquiring scientific degrees and holding academic positions.

II. Analysis of the candidate's career profile

Education and professional development:

2007 - 2013 - Higher Education "Pharmacy", Faculty of Pharmacy, Medical University - Plovdiv;
02-05.2012 - student exchange trainee within the ERASMUS program Radboud University Nijmegen, University Medical Center, Nijmegen (The Netherlands);
2013 - 2016 - Medical University - Plovdiv, Faculty of Pharmacy, Plovdiv under the supervision of Prof. Dr. Danka Obreshkova - Specialist in Analysis of Medicinal Products;
09-12.2014 - University of Namur, Namur (Belgium), specialization on international project "Development of FXIIa inhibitors a promising target to treat and prevent thrombotic diseases" (Prof. Lionel Pochet).

2013 - 2017 - PhD in Pharmacology (incl. Pharmacokinetics and Chemotherapy); Area 7. Health and Sport, professional field 7.1. Medicine - Medical University of Plovdiv, Faculty of Medicine, Department of Pharmacology and Clinical Pharmacology under the supervision of Prof. Dr. Damyanka Getova-Spasova;
2016 - 2018 - Assistant Professor, Medical University of Plovdiv, Faculty of Pharmacy, Department of Pharmacology and Drug Toxicology;
2018-2023 - Postdoctoral Researcher, Center of Plant Systems Biology and Biotechnology, Plovdiv;
From 2021 to present - Senior Assistant Professor, Bulgarian Academy of Sciences, Institute of Microbiology "Stefan Angelov", Department of Biotechnology, Laboratory of Metabolomics, Plovdiv (maternity break 08.2021-02.2023);
Since 2023 and currently - Coordinator for IMICB-BAS of the PlantaSYST project "Establishment of a Center for Plant Systems Biology and Biotechnology for the translation of basic research into sustainable bio-based technologies in Bulgaria".

Honors and awards:

- "Young Phytochemist" Award 2020 from the Bulgarian Phytochemical Association for achievements in pharmacological research related to the discovery of the potential of natural compounds as anti-obesity agents.
- Nomination for the Pythagoras Award for Young Scientist in Life Sciences and Medicine for 2023;
- Annual Award for Scientific Excellence of the "Stefan Angelov" Institute of Microbiology of the Bulgarian Academy of Sciences for 2023.

III. On the fulfilment of the scientometric criteria

Upon review of the submitted documents with the list of scientific works of Dr. Liliya Vladimirova Mihaylova (Vassileva), I found that her candidature fully meets the quantitative and measurable requirements of the IMicB - BAS for obtaining the academic position of Associate Professor, namely:

A. Scientific activity

1. Acquired educational and scientific degree "Doctor" - 2017. "Study of the effects of extracts of Rhodiola rosea L. and Curcuma longa L., as well as their active substances salidroside and curcumin on behavioral responses, cognitive and locomotor functions of rats with a model of chronic stress";
2. Actual publications beyond the PhD publications 37, as follows: 22 in foreign impact-factor journals, 2 with impact-rank; 13 in non-refereed journals and conference proceedings; first author of 13 of them;
3. Total impact factor of the articles - 130.96;
4. In addition, published in connection with the dissertation are 7 articles - 2 with impact factor, 1 with impact-rank, 4 in non-refereed journals and conference proceedings;
5. One publication in a university textbook;
6. Positive citations - 435 in total for the entire scientific career, which are available in the database of scientific information Scopus (author identification number 57191613000), excluding self-citations by the author and co-authors, accessed on 21.05.2024.
7. h-index - 12.

B. Scientific activity

1. Participation in scientific forums - 37 participations in international and national scientific conferences, of which 10 participations with oral reports and 26 with poster presentations, including those presented for the degree of Doctor of Education and Science;
2. Participation in scientific research projects - 8, of which 4 international (coordinator of the Bulgarian team in 1), and 4 national (leader of 1);
3. Bulgarian Phytochemical Association, Bulgarian Scientific Society of Pharmacy, European Phytochemical Association.

Total number of points for scientific activity - **6644.89** (minimum 400).

IV. On professional and teaching activities

1. Dr. Lilia Mihaylova has a total work experience in the specialty of over 10 years;
2. From 2016 to 2018 Dr. L. L. Mihaylova worked as an assistant professor at the Department of Pharmacology and Drug Toxicology, Faculty of Pharmacy, Medical University of Plovdiv; since 2021 she has been a senior assistant at the Department of Biotechnology, Laboratory of Metabolomics, Institute of Microbiology "Stefan Angelov", BAS.
3. Proficiency in a foreign language - English - excellent; German - basic.

V. Scientific contributions

The research interests of the chief assistant Dr. Mihaylova are focused on socially significant diseases and modern approaches to prevention and pharmacotherapy. Accordingly, the main scientific contributions of her work are in the field of molecular pharmacology of obesity, longevity and skin ageing with a focus on the mechanisms of action of natural substances of plant origin.

Her dissertation work was devoted to the study of pharmacological effects of 2 plant extracts - Rhodiola rosea L. (golden root) and Curcuma longa L. and their active components salidroside and curcumin and their combination in a rat model of chronic stress. The main contributions of the thesis and the materials published in connection with it are the beneficial effects of the studied plant extracts on body weight and proinflammatory cytokines, behavior, locomotor activity and spatial memory and orientation of rats subjected to chronic stress. Studies were conducted in healthy rats and rats with scopolamine-induced amnesia. The synergistic effects found are important. Of clinical contribution, it is concluded that standardized extracts of Rhodiola rosea L. and Curcuma longa L. may have potential benefit in the treatment of chronic stress and depression conditions in clinical settings due to adaptogenic, anti-stress, antidepressant and anti-inflammatory actions. The combination of the two molecules has promising potential in chronic stress and mild/moderate depression.

Dr. L. L. Mihaylova with a country-leading research team is very actively working on *preclinical models of obesity* in vitro in human adipocytes and in vivo in nematodes of the Caenorhabditis elegans species and on treatment modalities. Pioneering, theoretical-applied studies have focused on identifying plant secondary metabolites with activity against obesity and premature aging and determining their molecular mechanism of action. In addition, the possibility of synergistic interactions when incorporating natural molecules in combinations with drugs is being investigated. A screening platform for anti-adipogenic potential of plant extracts and natural molecules in an in vitro model of adipocyte differentiation of human adipose cells was developed. A system was implemented to assess anti-obesogenic potential at the organismal level in the nematode C. elegans.

Using these experimental setups, besides the possibility to evaluate the effects of a number of natural substances, light is also shed on the disruption of molecular signalling pathways in obesity. Thus, studies on the anti-obesogenic effect of Ononis spinosa L. extract and the pure substances ononin and mahakyain (marked beneficial effect) in a model of obesity of human adipocytes are of scientific and theoretical contribution. Rosmarinic acid treatment of human adipocytes showed marked anti-adipogenic and anti-inflammatory activity mediated by suppression of lipogenesis, activation of lipolysis, reduction in the levels of key transcription factors peroxisome proliferator-activated receptor gamma (PPARγ) and CCAAT/enhancer-binding protein alpha (C/EBPα); combined administration of caffeic and chlorogenic acids activates changes during differentiation of human adipocytes that induce so-called 'browning' of adipose tissue by activating AMPK and genes controlling mitochondrial metabolism.

Studies on the role of the inhibition of established signaling pathways in insulin resistance - that of phosphoinositide 3-kinase (PI3K)/protein kinase B (AKT) - should be noted as contributing to investigation of obesity in general and the possibilities of its treatment. In this aspect, a pronounced anti-adipogenic action and effect on PI3K/AKT signaling pathway of betulinic acid (quinap extract) in an in vitro model of human adipocyte obesity and regulation of insulin-mediated lipid accumulation in C. elegans subjected to a high-glucose diet. A mechanism of action has been proposed for an extract of Alchemilla monticola Opiz. and its secondary metabolites astragalin and quercitrin in a model of human adipocyte obesity. Tsaricha extract (A. monticola) markedly inhibits the PI3K/AKT signaling pathway in human adipocytes. An important finding is that such activity was not observed when the pure substances astragalin and quercitrin were administered alone.

Another important clinical and applied aspect of the contributions is the work on in vitro models *of skin aging and psoriasis* (in human keratinocytes). The anti-inflammatory potential and mechanism of action of plant culture extracts of Lavandula angustifolia L. and Harpagophytum procumbens (Burch) and the isolated pure substances rosmarinic acid and leucoseptoside A were studied in an in vitro model of psoriasis. Myconoside and calceolarioside E isolated from in vitro propagated Haberlea rhodopensis L. were found to exhibit photoprotective activity and prevented UV-induced oxidative stress damage to human keratinocytes by activating nuclear transcription factor E2-related factor 2 (NRF2).

Of practical importance in relation to the ageing population and the increasing role of anti-ageing therapies are studies in C. elegans on extracts and pure substances to influence *molecular mechanisms of longevity*. Icariin has been found to improve stress resistance and extend lifespan through a mechanism dependent on insulin-like signaling in nematodes. These data are extremely interesting in view of the leading role of insulin resistance in the aging process and tumorigenesis.

In the author's report Dr. L. Mihaylova outlines *future directions* in her scientific research activity based on her previous experience and current positions, which indicates future potential for academic development. In collaboration with the Department of Immunology at the IMicB-BAS, the CDFB and international partners, continuing studies on extracts and pure molecules in model systems of human adipocytes, keratinocytes, fibroblasts and C. Elegans and searching for activity against obesity, mitochondrial damage and UV-induced skin ageing, as well as potential for extending lifespan and reducing age-related metabolic disorders. In collaboration with IOHCF-BAS - combining different metabolomics platforms such as IMR, VETX combined with mass spectrometers to study the metabolome changes of plant extracts and model organisms (such as C. elegans) under different conditions - physiological, aging, stress and experimental treatments. Applied research in the field of pharmaceutical biotechnology in collaboration with the CDCB - biosynthesis, isolation and purification of economically relevant plant metabolites and development of biotechnological processes for the creation of new products for cosmetics and for the treatment of metabolic disorders and obesity.

VI. Conclusion:

The overall professional activity, the scientific experience and achievements and the realized theoretical and applied contributions of Dr. Lilia Vladimirova Mihaylova (Vasileva), give me grounds to give a positive opinion on her habilitation. The total number of points according to the NACID indicators is **7130.43.** I consider that her candidature **fully fulfils and exceeds the mandatory and additional conditions and scientific-metric criteria for holding the academic position of "ASSOCIATE PROFESSOR", which are in accordance with the requirements of the on the Development of the Academic Staff in the Republic of Bulgaria and the Regulations of the Institute of Microbiology at BAS.**

Dr. Liliya Vladimirova Mihaylova (Vassileva) has emerged as a hard-working, precise and thorough researcher with high scientific achievements and honors, a specialist-pharmacologist and lecturer with developed skills and good prospects for further academic development and enrichment of research and teaching activities of the Institute of Microbiology at BAS, Department of Biotechnology, Laboratory of Metabolomics, Plovdiv.

29.07.2024 г. Signed : .....
Plovdiv / prof. M. Orbetzova/