OPINION

from Prof. Dr. Pavletta Stoyanova Shestakova
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on the materials presented for the competition f
or the academic position of **"Associate Professor"**

at the Institute of Organic Chemistry with Centre of Phytochemistry (IOCCP), BAS
in the professional field **4.2. Chemical Sciences,** scientific specialty "**Organic chemistry"**

Senior Assistant Professor Dr. Neda Orlinova Anastassova from the Institute of Organic Chemistry with Centre of Phytochemistry, BAS, is the only candidate in the competition for the academic position of "Associate Professor" announced in the State Gazette, issue. 40 of May 16, 2025, and on the website of the IOCCP-BAS, for the needs of the Laboratory of Structural Organic Analysis at IOCCP-BAS.

1. General presentation of the materials and of the applicant

The set of materials presented by Dr. Neda Anastassova is in accordance with the Rules of the Development of the Academic staff of IOCCP-BAS and meets the criteria of IOCCP-BAS for the academic position of "Associate Professor".

Dr. Anastassova has presented 17 scientific papers for participation in the competition, which were all published in Impact Factor journals (Scopus and Web of Science) and are not included in her PhD Thesis. All 17 submitted publications are accepted for evaluation, 5 of which are under indicator B, 12 are under indicator G. The distribution of the journals on the respective quartiles (Q factors) is as follows: Q1 – 9, Q2 – 5, Q3 – 1, Q4 – 2.

Neda Anastassova graduated from the Master's Program "Fine Organic Synthesis" at the University of Chemical Technology and Metallurgy - Sofia (UCTM) and received the degree "Master of Chemistry" in 2012. In 2017, she was awarded the educational and scientific degree "Doctor of Pharmaceutical Chemistry" after successfully defending her PhD Thesis entitled: "Synthesis and study of hepatotoxicity and antioxidant activity of N,N'-disubstituted benzimidazole-2-thiones" with scientific supervisor Prof. Anelia Mavrova from UCTM, Sofia. During the periods January - February 2013 and April - September 2016, she worked as a chemist at the Institute of Organic Chemistry with Centre of Phytochemistry - BAS (IOCCP-BAS), from October 2016 she was appointed as an Assistant, and from May 2018 – as a Senior Assistant in the laboratory of SOA at the IOCCP - BAS. The overall scientific activities of Dr. Neda Anastassova are summarized in 26 publications with her participation, which have received 210 citations, excluding self-citations (reference in Scopus, H-index 9).

2. General characteristics of the applicant's activities

Evaluation of scientific and applied research activities

The scientific activity and scientific contributions of Dr. Anastassova are in the field of organic and pharmaceutical chemistry, with a focus on the development of new organic molecules and assessment of their potential as possible multitarget agents for the treatment of neurodegenerative diseases. This is an extremely relevant and socially significant scientific

topic, given the lack of effective therapeutic agents for the treatment of these types of diseases, their enormous social and economic scale, as well as their impact on the quality of life of patients and their families. Dr. Anastassova presents an extended overview of her scientific contributions, in which the conducted research is systematically and clearly discussed, and the main scientific achievements are described. The presented research covers the following main achievements: (i) Design, synthesis, spectral and structural studies of benzimidazole and indole hybrids; (ii) Study of the pharmacological activity of the compounds in various in vitro models; (iii) Study of the radical-scavenging properties of the compounds in various in vitro models; (iv) Conducting an in vivo study in a rat dementia model; (v) Quantum chemical calculations to elucidate the mechanism of action. The scientific contributions can be summarized as follows:

• 5 series of a total of 40 new arylhydrazone derivatives with different substituents were synthesized, including a combination of hydroxyl and methoxy substituents in the main molecular skeleton.

• Systematic and in-depth studies of their cytotoxicity (safety profile) were conducted using several in vitro models. The results allowed the identification of compounds with the most favorable safety profile and low cytotoxicity as preferred candidates for subsequent biological evaluation.

• A series of studies were conducted to evaluate the neuroprotective activity of some of the newly synthesized structures under oxidative stress conditions, as a key pathogenic mechanism in neurodegenerative diseases. For this purpose, a model of H_2O_2-induced oxidative stress in SH-SY5Y cells was used. Several compounds were identified that showed higher neuroprotective activity than that of the reference compounds (melatonin and rasagiline). The results obtained allowed the establishment of a relationship between the structure and biological activity of the compounds.

• For selected candidates, an additional assessment of the neuroprotective potential was performed using a model of induced neurotoxicity with 6-hydroxydopamine in isolated synaptosomes from rat brain. The results allowed to expand the neuroprotective profile of the obtained compounds.

• For series V, including arylhydrazones of indole-3-acetic acid, additional studies of neuroprotective activity were conducted with two additional models - a model of t-BuOOH-induced oxidative stress in isolated mitochondria from rat brain and in a model of oxidative stress in isolated microsomes, in which a combination of iron and ascorbic acid was used to induce lipid peroxidation.

• Based on the studies on neuroprotective activity, the relationship between the structure of the compounds and their effectiveness as neuroprotectors was clarified, and leading structures were identified, surpassing the effect of the reference compounds.

• For some of the compounds (Series III and IV), the potential permeability through the blood-brain barrier was studied, as a result of which a leading structure with high multitarget potential was identified, for which in vivo studies were conducted.

• Several structures were identified, selectively inhibiting the MAOB enzyme in the submicromolar range.

• Based on in vitro studies on the radical-scavenging properties of the synthesized compounds, conducted in systems with biologically significant molecules, as well as against

various free radicals, new knowledge was obtained about the structure-antioxidant activity relationship. The leading role of catechol fragments in the suppression of iron-induced lipid peroxidation, as well as for the antioxidant activity against various free radicals, was established.

• One of the compounds with the best complex performance, namely N'-(3,4-dihydroxybenzylidene)-5-methoxy-1H-indole-2-carbohydrazide, was studied in vivo in a rat model of scopolamine-induced Alzheimer's dementia. The effects on cognitive abilities and memory, the mechanisms of action and its toxicity were investigated. The results indicate that the compound has significant potential as a promising multi-target agent for the treatment of neurodegenerative diseases.

Dr. Anastassova's future plans are related to using the accumulated experience and knowledge for the synthesis of new multitarget compounds through rational design, aimed at a complex impact on the pathological mechanisms responsible for neurodegenerative diseases. It is planned to immobilize selected active molecules on nanoparticles, search for synergistic effects, as well as introduce new cell models (Parkinson's and Alzheimer's disease models) to assess the potential of new structures and systems. As a second area, the development of new antitumor agents including hybrid molecules and nanoparticles with potential synergistic action is planned.

Participation in projects and scientific forums, awards

Dr. Anastassova's research activity is realized through her active participation in research projects. Information is presented about participation in 13 research projects, and the large number of projects (5) of which she is the leader is impressive. The results of Dr. Anastassova's research have been presented at a significant number of international and national scientific forums, through scientific reports (19) and poster presentations (33).

The high level of Dr. Anastassova's research is recognized by the award from the National Competition "Acad. Yukhnovsky" for "Outstanding Young Scientist in the Field of Organic Chemistry" for 2018, as well as the awards for the best poster from three international scientific conferences.

Assessment of educational and pedagogical activity

Dr. Anastassova has significant experience in training students as a direct mentor of two young colleagues appointed at the IOCCP, she was a supervisor of one bachelor's and a co-supervisor of one master's thesis, mentor of three students under the Student Internship Program. This demonstrates the potential of Dr. Anastassova for attracting and training new young researchers.

Assessment of the applicant's personal contribution

Dr. Anastassova is the first author or the corresponding author in most of the publications included for participation in the competition. This is a recognition of her significant contribution to the conceptual design of the research and the realization of the scientific ideas. Therefore, I believe that her personal contribution to the publications submitted for participation in the competition is indisputable.

Personal impressions

My personal impressions of Dr. Neda Anastassova are wonderful, in my opinion she is one of the most talented and brilliant researchers of the younger generation at the IOCCP, with a markedly in-depth and intelligent approach to scientific work, demonstrating also independence, strong motivation and desire. I have been pleased to follow her scientific development over the years, and I am convinced that she has the potential and perspective to become one of the leading researchers in her scientific field, not only in Bulgaria, but also internationally.

3. Critical comments and recommendations

I have no critical comments on the work of Dr. Anastassova and the materials presented for participation in the competition. The documents are very carefully prepared, and the contributions are clearly and interestingly presented.

CONCLUSION

The scientific activity and research metric indicators of Dr. Neda Anastassova, reflected in the materials submitted for participation in the competition, cover and exceed the requirements for the academic position of "Associate Professor", in accordance with the Law for the Development of Academic Staff in the Republic of Bulgaria, The Regulations of BAS for the Implementation of this Law and the Regulations of IOCCP-BAS.

The scientific achievements of Dr. Neda Anastassova convincingly present her as a talented and promising scientist, distinguished by her own scientific profile and a thorough approach in topical and important scientific area.

After the analysis of the research output of Dr. Neda Anastassova, its importance and the scientific contributions reflected therein, **I give my positive assessment** and recommend to the Scientific Jury to prepare a report-proposal to the Scientific Board of IOCCP-BAS **for the selection of** Dr. Neda Orlinova Anastassova**, at the academic position of "Associate Professor" at IOCCP-BAS** in the professional field 4.2. Chemical Sciences, scientific specialty "Organic Chemistry".

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