

REVIEW

for competition for the occupation of the academic position "professor" in professional direction 4.2. Chemical sciences (Processes and apparatuses in chemical and biochemical technology) for the needs of the lab. "Biochemical Engineering" at the Institute of Engineering Chemistry (IEC) - BAS

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I am included in the scientific jury for the selection of an academic position "Professor" in the professional direction 4.2. Chemical sciences (Processes and apparatuses in chemical and biochemical technology) according to order ПД №15-467/26.09.2023 of the Director of IEC-BAS for the needs of the lab. "Biochemical Engineering" at the Institute of Engineering Chemistry (IEC) - BAS, announced in the State Gazette no. 67/04.08.2023. For participation in the competition documents are submitted by one candidate **Dr Daniela Ilieva Batovska**, chemist in IEC-BAS, which are in full agreement with requirements of the Development of Academic Staff in the Republic Bulgaria Act and the Regulations of its implementation.

1. Brief biographical data of the candidate.

Dr. Daniela Batovska graduated from NNMG "Acad. P. Chatalov" - Sofia in 1986, and in 1991 she graduated with a master's degree in organic and analytical chemistry at the Faculty of Chemistry (Faculty of Chemistry and Pharmacy) of SU "St. Kl. Ohridski" successfully defending a thesis on "Triterpenes and sterols in *Rosmarinus officinalis* L.". She started working as a chemist in the Department of Clinical Pharmacology and Therapeutics of the MU in December 1991, and in March 1993 she was appointed as a chemist in the lab. "Chemistry of natural compounds" of IOCCP - BAS. In the period 1997 - 2001, she was a self-taught doctoral student at IOCCP - BAS, where she prepared and successfully defended a dissertation on "Synthesis and biological activity of sterol derivatives" under the supervision of Prof. Tsenka Milkova. In the period 2000 - 2017, she successively held the positions of research assistant II and I and Associate Professor in the lab. "Chemistry of natural compounds" of IOCCP - BAS. In 2002, Dr Daniela Batovska won a two-year postdoctoral fellowship of Japan Society for the Promotion of Science (JSPS), which she carried out at the Biotechnology Research Center, Toyama Prefectural University and the Graduate School of Agriculture, Hokkaido University, Sapporo, Japan. In the period 2009 - 2010, Dr. Daniela Batovska also worked as a part-time teacher at the SWU "Neofit Rilski" - Blagoevgrad, where she led lectures and exercises on Steroid Chemistry and Bioorganic Chemistry. In the period 2017 - July 2022, she was a chemist at RLG 2016 OOD - Sofia, whose main activity was the development of food and cosmetic products. Since August 1, 2023, she has been a chemist in the lab. "Biochemical Engineering" at IEC - BAS.

2. General characteristics of the scientific research and scientific applied activity of the candidate

The main research activities of Dr Daniela Batovska are in the following directions:

- synthesis of biologically active chalcones and study of the relationships between their chemical structure and their biological activity - green synthesis and synthesis of chalcones with antioxidant and/or antimalarial activity, or as inhibitors of the overexpression of glycoproteins encoded by the MDR gene in tumor cells;
- development and adaptation of spectrophotometric methods for the determination of antiradical activity in the microscale;
- phytochemical studies of medicinal plants such as *xGraptoveria*, *Potentilla reptans*, *Lavandula angustifolia*, etc. and analysis of their products;

The scientific topic on which Dr. Daniela Batovska works is complex and interdisciplinary. This is also evident from the publications of Dr. Batovska, in which scientists from various fields - chemistry, biology, biochemistry, microbiology, medicine, etc. are present. The impressive number of citations noticed so far in Scopus/WoS (1428) is a vivid confirmation of the significance of the obtained results and the relevance of the topic on which Dr. Batovska works.

Dr Batovska is the leader of two international scientific projects financed by the Scientific Research Fund - with Germany and India and is the supervisor of a successfully defended doctoral student from India.

In recent years, Dr Batovska gained valuable experience in the field of developing food and cosmetic products based on natural products, moving from the purely scientific area to the private business sector. This experience, combined with the previous scientific and research activity, define Dr Batovska as a completely suitable candidate for the announced competition for the purposes of the lab. "Biochemical Engineering" at IEC - BAS.

3. Evaluation of the presented materials.

The results of the research activity of Dr Daniela Batovska are reflected in a total of 60 scientific publications, of which 47 are peer-reviewed in the world databases Scopus/WoS, with a H-index of 16. To participate in the current competition, the candidate submitted 20 scientific works (other than those included in competitions for PhD and associate professor), 1 chapter of book and 1 patent, with which the candidate fulfills and exceeds the minimum national requirements according to the regulations for the implementation of national requirements under the Development of Academic Staff in the Republic Bulgaria Act and the Regulations of the BAS, as follows:

According to group of indicators A - 50 points (required 50 points): PhD Dissertation work on the topic "Synthesis and biological activity of sterol derivatives" (2001) in the scientific specialty: 01.05.10 "Bioorganic chemistry, Chemistry of Natural and Physiologically Active Substances" in IOOCCP - BAS.

According to group of indicators B (habilitation work) - 120 points (required 100 points): A total of 6 publications (2-Q1, 2-Q2 and 2-Q3) are presented in scientific journals, referenced and indexed in the world-famous databases Scopus/ WoS.

According to group of indicators G – 274 points (required 220 points): 16 publications in journals, referenced and indexed in world-renowned databases (1-Q1, 10-Q2, 3-Q3, 2-Q4 and 1 without Q/SJR) are presented, as well as 1 book chapter. Dr D. Batovska is also a co-author of a patent (US 2019/0390408 A1, WO2020044343,05 Mar 2020).

According to group of indicators D – 776 points (required 120 points): The number of citations of Dr D. Batovska's scientific publications (383), included in the competition for "professor" position, which are available in the scientific information databases Scopus/WoS, is impressive.

According to group of indicators E – 160 points (required 150 points): In this group, the candidate has accumulated 160 points as the leader of a Bulgarian team in international scientific projects (100 points), the supervisor of a successfully defended doctoral student (50 points) and funds attracted from projects (10 points).

The total number of points from all indicators is 1370. Dr D. Batovska significantly exceeds the additional criteria for academic growth according to the Regulations of IEC-BAS in terms of total number of publications (60 with a minimum of 40), number of publications presented in the competition with an impact factor /impact rank (19 with a minimum of 7), citations (383 with a minimum of 50) and H-index (16 with a minimum of 8).

4. Main scientific and scientific-applied contributions.

The main scientific contributions of Dr Daniela Batovska in the field of design, synthesis, structural determination and biological activity of chalcones are:

1. Literature data on the pharmacological activity of natural and synthetic chalcones are summarized (publ. № 1).
2. Chalcones were obtained for the first time by an enzyme-catalyzed Claisen-Schmidt condensation between benzaldehyde and acetophenone, catalyzed by lipase from porcine pancreas and in the presence of imidazole as a promoter, as with acylase from *Aspergillus melleus* (3.5.1.14). It was found that the latter, as well as the recombinant D-aminoacylase, also catalyze the reaction between acetophenone and p-nitrobenzaldehyde (publ. № 3).

3. Sixteen hydroxychalcones were synthesized and characterized, and a relationship between structure and scavenging ability towards the stable DPPH radical was established (publ. № 3). Using a highly sensitive chemiluminescence method, the interaction of 6 of the chalcones with peroxide radicals was investigated (publ. № 2). The study of the mechanism of antioxidant action of chalcones showed that they also exhibit a pro-oxidant effect as part of the radicals formed by them react with molecular oxygen to form unstable 1,2-dioxetanes (publ № 7).

4. Twenty-four chalcones with a different alkylation pattern in ring A were synthesized and structurally characterized. Of them, 10 chalcones were found to inhibit schizont maturation with IC_{50} in the range 0.10-0.55 $\mu\text{g/mL}$ for two strains of *P. falciparum*, one susceptible, and the other resistant to chloroquine (publ. № 4). 3,4,3',4',5'-Pentamethoxychalcone (IC_{50} 0.11 mg/ml) was also found to possess 13-fold higher activity than licochalcone A and a high selectivity index. The three most active compounds were found to cause significant disruption of all parasite membranes, including those of the nucleus, mitochondria and food vacuole. (publ. № 5). The *in vivo* pharmacokinetic study in New Zealand white rabbits showed a severe limitation of the therapeutic efficacy of the three chalcones against tropical malaria due to their low bioavailability, which is most likely due to the targeting of the chalcones to other targets, e.g. to P-glycoprotein (publ. № 6). Also, in an *in vivo* human tropical malaria study model, the three chalcones were found to modulate the immune response of infected mice and strongly decrease interleukin 12 levels (publ. № 14). In another *in vivo* model of mouse with induced cerebral malaria, a significant reduction in percent parasitaemia was observed at day 10 post-infection (publ. № 19). The obtained results indicate that chalcones with 3',4',5'-trimethoxy- and 2',5'-dimethoxy-substitution patterns in ring A are promising antimalarial agents, but the substituents in ring B need to be carefully considered.

5. Based on 3 previously established leader structures, 15 chalcones were synthesized and evaluated for reversibility of multidrug resistance (MDR) in murine lymphoma cells (publ. № 8). The most active chalcones were found to be stronger revertants than the positive control, verapamil. In a combination chemotherapy model, one of the most active compounds, 4-dimethylamino-2',4'-dimethoxychalcone, was shown to have an additive effect when interacting with the anticancer drug doxorubicin. Experimental results as well as quantum chemical calculations show that the substituents in the p-position of both rings should have a hydrophobic nature.

6. Twelve chalcones were synthesized by Claisen-Schmidt aldol condensation between substituted acetophenones and aromatic aldehydes. The study of the antibacterial activity of these chalcones against 3 types of bacteria – *Staphylococcus aureus*, *Streptococcus pyogenes* and *Corynebacterium diphtheria* shows that their activity depends on the presence of the terminal methoxyl groups in the structure of the chalcones (publ. no. 20).

In the field of phytochemical research, the following more important scientific contributions can be mentioned:

1. Adapted spectrophotometric methods developed with the Elisa reader were developed in order to determine antiradical activity against highly reactive oxygen- and nitrogen-containing particles present in living systems as: super oxide anion radical ($O_2^{\bullet-}$), hydroxyl radical (OH^{\bullet}), hydrogen peroxide (H_2O_2) and nitric oxide (NO^{\bullet}) to determine the activity of methanolic extracts of aerial and root parts of *in vitro* cultivated *Artemisia alba* and aerial parts of *in vitro* cultivated *Clinopodium vulgare*, *Hypericum tetrapterum*, *H. richeri* and the endemic *H. rumeliacum* to NO^{\bullet} (publ. № 9) and to synthetic compounds as analogs of polyhydroxylated trans-blocked 2-aryl cinnamic acids to OH^{\bullet} and $O_2^{\bullet-}$ (publ. № 11) and of thiazolo- and isoxazolo-hydrazinylidene-chromane-2,4-diones to $O_2^{\bullet-}$ and NO^{\bullet} (publ. № 12).

2. The chemical composition of fresh juice obtained from the leaves of the succulent plant *xGraptopetia* (Crassulaceae) (publ. № 10) was investigated by GC/MS analysis. Alkylamines, hydroxycarboxylic acids, aliphatic and aromatic carboxylic acids, amino acids, alcohols, aromatic and aliphatic hydrocarbons have been identified.

3. The antibacterial activity against 3 strains of *S. aureus* of a water-alcoholic extract of *Potentilla reptans* and its fractions was investigated. The hexane fraction was found to have the best bacteriostatic activity (publ. № 13).

4. The chemical composition of essential oil from *Lavandula angustifolia* (lavender) variety Sevtopolis, grown in an agricultural plantation near the city of Pomorie, was investigated by GC/MS analysis (publ. № 17). Forty-four components were identified with main ingredients linalool (24.1%) and linalyl acetate (27.5%), which serve to evaluate the quality of the essential oil. It was found that in terms of its main components, Bulgarian lavender oil meets the standards set by ISO (2002) and by the European Pharmacopoeia (10th edition, Council of Europe 2020).

5. The review of the literature data on gentians (Gentianaceae) as pain control (publ. № 15) and wound healing (publ. № 16) and secondary metabolites of gentian species from 5 sections growing in the Bulgarian mountains (publ. № 18), which will facilitate the finding of compounds responsible for the field-observed susceptibility of some of the gentian species to certain insect invaders.

Some of the presented results, such as those related to antioxidant and antimalarial activity, also have a scientific-applied contribution and have a high potential for practical application.

The presented materials show that Dr Daniela Batovska has a thorough and excellent knowledge of the state of research in the field of synthesis and design of new chalcones, of modern methods for their characterization. It is noteworthy that she has mastered the specific microbiological and biochemical terminology and skilfully analyzes the obtained results, both in the field of organic synthesis and in the field of microbiology and medicine. I believe that Dr

Batovska with her knowledge and experience has made a significant contribution to the presented publications, and the formulated contributions and obtained results are largely her personal work..

5. Reflection of the candidate's scientific publications in Bulgarian and foreign literature.

Dr Daniela Batovska has provided a list with 383 citations of the publications included in the competition. It is worth to mention, the impressive number of citations of publication No. 1 – 261, which is indisputable proof of the topicality of the topic on which Dr Batovska works.

6. Critical notes and recommendations.

I have no critical comments on the presented documents. All results have been published in peer-reviewed international journals and have passed the critical evaluation of experts in the respective fields.

7. The reviewer's personal impressions of the applicant.

I know Dr Daniela Batovska personally and have excellent impressions of her professional and collegial qualities. Her capacity for work, determination and dedication to science are remarkable.

CONCLUSION

The documents and materials presented by Dr Daniela Batovska, meet all the requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria (ZRASRB), the Regulations for the Implementation of ZRASRB, the relevant Regulations of BAS and the specific requirements of the Regulations of IEC-BAS.

From the presented habilitation report and scientific publications, both original scientific and scientific-applied contributions can be clearly seen, proving that the candidate is a built scientist with interdisciplinary qualifications and experience. All this gives me reason to give my positive assessment and to recommend to the Scientific Jury to prepare a report-proposal to the Scientific Council of IEC-BAS for the election of Dr Daniela Batovska to the academic position of "professor" in professional direction 4.2. Chemical sciences (Processes and apparatuses in chemical and biochemical technology) for the needs of the lab. "Biochemical Engineering" at the Institute of Engineering Chemistry (IEC) - BAS.

09.11.2023

Reviewer:



(Prof. Dr Antoaneta Trendafilova)