REVIEW

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on the materials submitted for participation in the contest for academic position "Associate professor" in: higher education field 4. Natural Sciences, Mathematics and Informatics; Professional field 4.2. Chemical sciences (Organic Chemistry, Chromatographic analysis)

In the contest for "Associate professor" announced in state newspaper (no. 39 of 02.05.2023) and on the official website of Plovdiv University "Paisii Hilendarski" for the needs of the Department of Organic Chemistry at the Faculty of Chemistry as the only candidate participates chief assistant professor Dimitar Georgiev Bozhilov, PhD, from the same department.

General presentation of the received materials

According to order № РД-21-1425 from 30.06.2023 г. of the Rector of Plovdiv University "Paisii Hilendarski", I have been approved as a member of the scientific jury for the procedure for academic position "Associate professor" in: higher education field 4. Natural Sciences, Mathematics and Informatics; Professional field 4.2. Chemical sciences (Organic Chemistry, Chromatographic analysis) announced for the requests of the Department of Organic Chemistry at the Faculty of Chemistry.

As the only candidate with submitted documents is chief assistant professor Dimitar Georgiev Bozhilov, PhD from the Department of Organic Chemistry at the Faculty of Chemistry in Plovdiv Plovdiv University "Paisii Hilendarski".

The presented by doctor Dimitar Bozhilov set of documents are in accordance with the Regulations for development of the academic staff of the Plovdiv University "Paisii Hilendarski" meets the criteria and includes documents as follows: Application; CV; Diplom for Master drgree; Diplom for PhD degree, List and copies of scientific works; List of citations; Reference with compliance with the minimum national requirements; Annotation of materials with extended habilitation reference; Self-assessment of contriburions; Declaration of originality and authenticity etc.

The candidate Dr. Bozhilov presented a total of 46 scientific papers, 11 of which were published in journals referenced outside of Web of Science and Scopus, another 11 are full-text papers in conference proceedings. Twentyfour (24) scientific works that are outside the dissertation are accepted for review, have scientometrics and are reported in the final evaluation, 10 publications without scientometrics on the subject of the competition, 11 reports

in collections and 14 research projects. Three scientific papers on the dissertation are not reviewed. The reference for the fulfillment of the minimum requirements covers a total of 24 publications corresponding to the scientific specialty of the competition. The presented materials are well designed and clearly present scientific activity and achievements of the candidate.

Chief assistant professor Dimitar Bozhilov, PhD, participated in the competition with a total of 24 scientific publications, distributed according to the rank of the scientific journals, expressed in quartiles (Q-factor), as follows: 6 publications in scientific journals with Q1; 2 publications in scientific journals with Q2; 4 publications in scientific journals with Q3; 14 publications in scientific journals with Q4 (7 of them in refereed editions with SJR without IF).

Seven (7) of the publications are presented under **group B** of the indicators (referenced to a Habilitation thesis) with a total of 155 points. The rest 17 of the publications are presented under **group G** of the indicators.

Brief biographical data

Ch. ass. Prof. Dimitar Bozhilov, PhD completed his higher education at the Faculty of Chemistry of the PU, where in 2008 he obtained a bachelor's degree in “Computational chemistry”, and in 2009 - a master's degree in "Organic chemistry". In 2017, he defended his PhD dissertation on the topic: “Phytochemical studies on the essential oil and polyphenolic complex of yarrow (Achillea Millefolium), peppermint (Mentha Piperita) and fireweed plant (Chenopodium Botrys L.)“, under the supervising of Assoc. prof. Soleya Dagnon, PhD in the department of organic chemistry in Plovdiv university. Dimitar Bozhilov began his scientific activity at the PU as a full-time PhD student in Organic Chemistry in 2010, subsequently as assistant professor (2013-2018) and chief assistant professor (from 2018 to present) in the same department.

General characteristics and activity of the candidate

Educational and pedagogical activity of Dr. Bozhilov is diverse and intense and covers:

1. Lectures and lab exercises in Bioorganic chemistry for bachelors;
2. Lectures and lab exercises in Modern chromatographic methods for bachelors;
3. Lectures and lab exercises in Chromatographic analysis for the bachelor’s program "Analysis and control";
4. Lectures and lab exercises in Analysis of medicinal substances for the bachelor’s program "Medicinal chemistry";
5. Lectures and lab exercises in Chromatographic methods in the pharmaceutical analysis of students from Master's programs of the Faculty of chemistry at Plovdiv university;

6. Laboratory and seminar exercises in Organic Chemistry;
7. Lectures, laboratory and seminar exercises on Mass Spectrometry and UHPLC-MS for undergraduate and graduate students, as well as PhD students.

His annual workload for the period 2018 - 2023 (with a 360-hour plan) is between 403 and 584 hours per year, or a total of **2428** hours for the period. The candidate is author of study programs in two disciplines from the Master's program Pharmaceutical chemistry, as follows: Chromatographic methods in pharmaceutical analysis and Phytochemistry of medicinal plants.

Dr. Dimitar Bozhilov is the head of the postgraduate program on High Performance Liquid Chromatography (HPLC), leading specialized courses in HPLC-DAD-MS for bachelor and master graduates. In addition, supervising of diplom work with 13 students from bachelor's and master's programs for the period 2019-2022, as well as the contribution of students in research projects. All of this presents the candidate as a well-established academic teacher with interests and competencies in the field mainly of Chromatographic analysis, but also in Bioorganic chemistry and Pharmaceutical analysis.

In the **scientific and scientific-applied activity** of Dr. Bozhilov can be summarized three directions: the main part of the research covers the application of chromatographic methods and techniques for the analysis and identification of secondary plant metabolites (components of the polyphenolic complex, as well as components of the essential oil composition); *in vitro* spectrochemical methods and techniques for studying biological and physiological activity of various extracts and pure substances; structural characterization of new bioactive compounds. The polyphenolic complex of *Habarlea rhodopensis*, *Chenopodium botrys*, *Vernonia amygdalina*, *Rosa damascena Lactuca spp – Lactuca sativa Batavia cv*, *Maritima*, *Lolo rosa cv. Tuska* and *cv. Winter Butterhead* was investigated. This includes analysis, identification, quantification of the main polyphenolic components and *in vitro* biological activity examination. The essential oil composition of *Nepeta transcaucasica* and *Nepeta cataria* was investigated. In addition, the antioxidant activity of the essential oils of *Helichrysum italicum* and two types of *Nepeta spp* was examined.

The extended habilitation reference correctly summarizes the achievements and contributions presented in the designated seven publications (replacing a habilitation thesis), published in the period 2017-2023. Five (5) of them are published in highly rated journals with Q1 rank: Plant Physiology and Biochemistry (2017, IF 2.718), Journal of Food Composition and Analysis (2019, IF 3.721), Turkish Journal of Agriculture and Forestry (2020, IF 2.585), ACS Omega (2023, IF 4.132), Molecules (2023, IF 4.927). The remaining 2 publications are in peer-reviewed journals with Q3 rank: Czech Journal of Food Sciences (2020, IF 1.279) and Bulgarian Journal of Agricultural Science (2021, no IF, SJR 0.216). The total Impact factor (IF)

of these articles is 19.353, the average is 2.763.

The main contributions:

The investigations have **scientific** and scientific-applied contributions in the field of chromatographic analysis and the methodology of GC-MS, HPLC-PDA, HPLC-MS/MS, UHPLC-MS/MS, for identification and explanation of the structure of biologically active compounds. 6 new gallic acid glycosides in the polyphenolic composition of *Rosa damascenesa* have been identified, from them quercetin galloyl hexoside and kaempferol disaccharide are reported for the first time. In 2023, three new compounds are identified in the composition of *C. botrys* - pectolinarigenin, sinensetin isomer and demethylnobiletin, which have not been reported for this plant so far. 19 polyphenolic compounds in *Vernonia amygdalina* are identified using surrogate materials, such as caffeoylquinic acids, apigenin and luteolin glycosides. Methods for obtaining of unique chromatographic profiles "fingerprints" have been developed containing a maximum number of well-separated peaks using Purospher, Kromasil, Sinergi and Nucleosil RP C18 chromatographic columns and mobile phases with the addition of formic and trifluoroacetic acids (Georgieva et al. 2017; Dagnon et al. 2019; Bojilov et al. 2020, 2023; Dragoev et al. 2021).

The examination of the antioxidant activity from essential oils of *Helichrysum iatalicum* and *Nepeta spp* using ABTS, DPPH, CUPRAC and FRAP methods introduces **scientific and applied** contribution. The essential oils from *H. italicum* and *N. cataria* are characterized and shows high antioxidant activity due to their high content of oxygen containing monoterpenes. The chemical composition, antimicrobial and antioxidant potential of essential oils from two subspecies of *Helichrysum italicum (Roth) G. Don* grown in Bulgaria were also investigated. Their composition and antimicrobial activity were compared with essential oils originating in Bosnia and France. In this regard, these results (Mollova et al. 2020, 2023) have had 28 citations in articles referenced in Scopus noted so far.

The polyphenolic profiles (fingerprints) of the resurrection plant *Haberlea rhodopensis* were obtained for the first time including all components of the complex. It was found that the polyphenolic complex of *H. rhodopensis* includes only two types of glycosides - phenylethanoid glycosides and hispidulin 8-C-glycosides. The main role of phenylethanoid glycosides for the protection of *H. Rhodopensis* has been demonstrated. On the article related to these results, 20 citations found in Scopus are observed by the end of May 2023.

An analytical approach for the determination of polyphenolic components in *Vernonia amygdalina Del.* using surrogate standards is proposed, extracts of green coffee and *Achillea asplenifolia* 9602 and rutin as an internal standard. HPLC–PDA fingerprint profiles of the polyphenolic complex of Vernonia amygdalina and surrogate standards were developed. The presence of luteolin 4′-O-glucoside, apigenin 7-O-rutinoside, apigenin 7-O-glucoside and

apigenin as minor constituents in *Vernonia amygdalina* is reported for the first time. The results suggest the application of the approach with surrogate standard in food analytical practice as highly recommended. The publication is cited in seven refereed journals.

Moreover, an extract of *C. Botrys* containing polyphenols is fractionated for the first time. The composition of the fractions is quantitatively determined and *in vitro* studies of their biological activity are carried out. The fractions exhibit antioxidant activity assessed by HOSA, HRSA and NOSA. Furthermore, Quercetin diglycosides are found to exhibit affinity for scavenging ROS, while 6-methoxy flavones are more active towards neutralizing nitroxide radicals. Quercetin glycosides and 6-methoxy flavones are characterized by high *in vitro* anti-arthritic activity assessed by ATA, but only mono- and diglycosides demonstrate high *in vitro* anti-inflammatory activity. It is suggested that these classes of compounds may find application in the development of new pharmaceutical products with medical and pharmacological applications.

The results of the study of antioxidant activity of extracts of seven types of wild growing edible mushrooms (*Boletus pinophilus*, *Cantharellus aurora*, *Cantharellus tubaeformis*, *Cantharellus cibarius*, *Craterellus cornucopioides*, *Morchella esculenta* and *Tricholoma equestre*) collected in the Rhodopes in the region of the city of Batak are of applied importance. DPPH, ABTS, FRAP and CUPRAC were used as suitable methods to evaluate their antioxidant activity. *Boletus pinophilus* is found to be characterized by the highest activity. The contributions from the studies of *in vitro* biological activity of newly synthesized biofunctional hybrid molecules have also an applied character. The antioxidant activity is evaluated by hydrogen peroxide deactivation (HPSA). Compounds containing a profene fragment in their structure are studied for *in vitro* anti-inflammatory activity (by inhibiting the denaturation of albumin – IAD) and *in vitro* anti-arthritic activity (anti-tryptic activity – ATA). Moreover, all hybrid molecules are found to inherit the anti-inflammatory and anti-arthritic properties of profenes. Newly synthesized tetrahydroisoquinoline sulfonamides are characterized by significant antioxidant (HPSA), *in vitro* anti-inflammatory activity (IAD) and anti-arthritic activity (ATA).

The originality of the research and the personal involvement of Dr. Bozhilov are indisputable. In 3 of the articles the candidate is the first author, and in the remaining 21 articles he is the third and subsequent author. This is reasonable due to the fact that most of the investigations are interdisciplinary and combine at least two scientific fields. The largest number of publications (13) are in journals from MDPI, as follows: Molbank - 9, Molecules - 2 and Processes - 2. Another 5 of the articles are published in Bulgarian journals, as follows: Bulgarian Chemical Communications - 3, Oxidation Communications - 1 and Bulgarian Journal of Agricultural Science - 1. The remaining 6 publications are in various reputable journals in the fields of chemistry, biochemistry, food and agricultural sciences. The total IF of the articles

which Dr. Bozhilov is participated included for the competition is 34,893.

The results of the research are widely appreciated in the works of many foreign authors. From the total of 46 publications, submitted for participation in the competition and the publications included in the PhD dissertation, a total of 130 citations of 24 articles are observed. More than 100 of these citations are in refereed and indexed journals and bring 206 scientometric points, but also there are citations in dissertations (8) and diploma theses (3) of foreign authors. Only on the articles representing habilitation work, 56 citations are noticed in refereed journals. Considering, that two of them are published in 2023 in open access journals (one already cited), many more citations are expected. The scientific response confirms the scientific-applied character of the research and the significance of the contributions. The presence of citations in refereed journals (16) of articles published in non-refereed ones makes a great impression. Therefore, this fact is proof of the quality of the developments and the applicability of the results. At the same time, only seven of the observed citations in refereed journals are on research in the synthetic field.

The quantitative indicators according to the criteria of the national requirements for an academic position "Associate professor" are met or exceeded - indicator **B** by more than 50% and indicator **D** by 300%. The specific requirements of the Faculty of Chemistry for a three-year full-time teaching experience are also significantly exceeded - the candidate Dr. Bozhilov has ten years of teaching experience, five years as chief assistant professor.

Personal impressions

I have known Dimitar Bozhilov since he was a student and I have observed his growth and academic career. The qualities of accuracy, persistence and purposefulness were complemented over the years by thoroughness and analyticalness.

CONCLUSION

The presented by chief assistant professor Dimitar Georgiev Bozhilov, PhD set of documents **are in accordance** with the Regulations for Development of Academic Staff in the Republic of Bulgaria Act (DASRBA), the implementing regulations of (DASRBA) and the relevant regulations of the PU "Paisii Hilendarski".

The candidate in the competition is submitted a **significant** number of scientific works published after the materials used for the defense of "Doctoral" degree and chief assistant. In this regard, there are original scientific and applied contributions that have received international recognition. Most of them are published in journals from international academic publishers. His research has practical application, and the main part of them are directly oriented towards academic work. Dr. Bozhilov's scientific and teaching qualifications **are unquestionable**. The

educational and research achievements of chief assistant professor Dr. Dimitar Georgiev Bozhilov, PhD **completely correspond** with the Regulations for development of the academic staff of the Plovdiv University "Paisii Hilendarski".

Based on the above, after examining the materials and scientific works presented in the competition, analysis of their significance, including scientific and applied contributions, I find it reasonable to give my **positive assessment and recommendation** to the esteemed academic board to prepare a report-proposal to the Faculty Council of the Faculty of Chemistry and recommend Dimitar Georgiev Bozhilov for academic position "Associate professor" at Plovdiv university „Paisii Hilendarski“ in: higher education field 4. Natural Sciences, Mathematics and Informatics; Professional field 4.2. Chemical sciences (Organic Chemistry, Chromatographic analysis).

01.09.2023 Reviewer:

Plovdiv Assoc. Prof. Stela Statkova-Abeghe