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of the materials submitted for participation in a competition for the academic position of 'Associate Professor' at the Institute of Botany and Ecosystem Research (IBER), Bulgarian Academy of Sciences in the field of higher education: 4. "Natural Sciences, Mathematics and Informatics"; Professional field: 4.3. "Biological Sciences"; Scientific specialty: "Botany"

1. General presentation of the received materials

In the competition for "associate professor", announced in the State Gazette, issue 66 of 06.08.2024 and on the website of IBER, BAS, Dr. Boryana Sidzhimova, Assist. Prof. at IBER, BAS, participated as the only candidate. The set of materials submitted by Dr. Boryana Sidzhimova is in accordance with the Regulations for the development of the academic staff of IBER, and meets the criteria of IBER-BAS for occupying the academic position of "Associate Professor". A certificate of fulfillment of the minimum requirements, a certificate of scientific contributions, a list and copies of scientific papers for the competition (a total of 23, corresponding to the scientific specialty of the competition), as well as a certificate of work experience, a diploma for acquiring PhD degree and a list of the candidate's teaching activities are attached. The submitted materials are well-structured and clearly present the candidate's previous scientific work and achievements.

2. Brief biographical data

Dr. Boryana Sidzhimova graduated from the Master's degree program in Botany in 2002 at the Faculty of Biology of Sofia University "St. Kl. Ohridski", Sofia. In 2001 she was appointed as a biologist at the Institute of Botany, BAS, and in 2004 she was enrolled in a full-time PhD program at the same Institute. In 2008 she successfully defended her dissertation thesis for obtaining the educational and scientific degree "Doctor" in the scientific specialty 01.06.03 "Botany" at IB (IBER)-BAS. From 2007 to 2011 she held the position of "biologist", and from 2013 until now - the academic position of Assistant Professor at IBER-BAS. Dr. Boryana Sidjimova is a participant in more than 20 projects with national and international funding. The results of her research for the last 4 years have been the subject of a total of 15 publications in refereed scientific journals. Dr. Boryana Sidzhimova teaches classes and summer practices to students of the Faculty of Biology of Sofia University "St. Kl. Ohridski". She is a member of the Bulgarian Botanical Society.

3. Fulfillment of the requirements for the academic position of Associate Professor

For participation in the present competition Dr. Boryana Sidjimova has submitted a report on the fulfilment of the minimum requirements for holding the academic position of Associate Professor at IBER-BAS as follows: **Group A - 50 points** (required **50** points): Dissertation entitled "Biological and phytochemical study of the genus *Galanthus* L. (snowdrop) in Bulgaria" for obtaining the PhD degree in the scientific specialty 01.06.03 Botany, IBER-BAS.

Indicator 4 in Group B - 110 points (required 100 points): A total of 7 publications equivalent to a habilitation thesis have been submitted (1-Q1, 1-Q2, 3-Q3, 1-Q1 without IF and 1-Q4 without IF) (*Biochemical Systematics and Ecology, Chemistry & Biodiversity, Heliyon, Ecologia Balkanica, Industrial Crops & Products, Z. Naturforsch.(C)* and *Compt. Rend. Acad. Bulg. Sci.*).

Indicators 7 and 8 in group D - 250 points (required 220 points): Submitted 15 publications in journals refereed and indexed in world-renowned databases with IF and/or SJR (2-Q1, 3-Q2, 5-Q3, and 5- SJR). (*Biotechnology and Biotechnological Equipment, Ecologia Balkanica, Caryologia, Biologia, Rapid Communications in Mass Spectrometry, South African Journal of Botany, Bio Risk, Planta, Acta Microbiologica Bulgarica, Journal of Plant Physiology and Compt. Rend. Acad. Bulg. Sci.*), and 1 book chapter.

Indicator 12 in Group E - 84 points (required **60** points): The number of citations of the scientific publications of Assist. Prof. B. Sidzhimova, included in the competition for associate professor, which are available in the scientific information database Web of Science and/or Scopus (with IF and/or SJR) is 42.

The total number of points from all indicators is **494**, with which Dr. Sidzhimova exceeds the minimum requirements for the academic position of Associate Professor according to the Regulations of IBER-BAS.

4. Evaluation of the candidate's scientific and applied scientific activity

The species of the subfamily Amaryllidoideae of the family Amaryllidaceae, including 59 genera and 850 species, contain a specific type of alkaloids, known as Amaryllidaceae alkaloids, which have diverse pharmacological activity. The most famous representative of this group of compounds is galantamine, isolated for the first time from *G. woronowii*, which is a very good inhibitor of acetylcholinesterase and is used to treat mild and severe forms of Alzheimer's disease. The topic is particularly relevant today, when over 7 million new cases of dementia are registered worldwide every year. The search for new sources of galantamine and other Amaryllidaceae alkaloids, the establishment of new biological activities, the correct taxonomic determination of species, the possibilities for cultivating species rich in this type of alkaloids are the main directions on which intensive work has been carried out over the past 20 years worldwide. The scientific research of Assistant Professor Dr. Boryana Sidzhimova is also in this direction, namely studies of species of the genus *Galanthus* and other representatives of the Amaryllidaceae family (*Narcissus* cv. *Hawera*, *Narcissus pallidulus*, *Hippeastrum papilio* (Ravenna) Van Scheepen), as well as on other plant species such as *Tribulus terrestris* L., *Colchicum autumnale* L., *Rhodiola rosea* L., representatives of Asteraceae and Lamiaceae, etc.

The activity of Assistant Professor Dr. Boryana Sidzhimova is interdisciplinary, including research from various scientific fields such as phytochemical studies (mainly alkaloid composition) and chemotaxonomic studies, botanical (anatomical, comparative-embryological, genetic and morphological) studies and taxonomic solutions, biological activity, cultivation experiments, as well as distribution and monitoring of plant species.

The most important scientific contributions from phytochemical research are:

- For the first time, a significant number of populations of *G. nivalis* and *G. elwesii* of Bulgarian origin, as well as sympatric populations, were studied and the accumulation of alkaloids in the individual organs (roots, bulbs, leaves and flowers) of both species was monitored, identifying 70 alkaloids of the galantamine, lycorine, homolycorine, tazetine, hemantamine, narcyclazine and tyramine types, and establishing intraspecific diversity in the alkaloid profiles with respect to the main types of alkaloids (chemotypes) (publ. B2).

- In a study of *G. elwesii* and *G. nivalis* with the same habitat over 20 alkaloids were identified of which, only 5 are common to both species and ismine, hamaine, carrageenan, incartine and 6-deoxytazethine are new to the genus *Galanthus* (publ. B1).

- It has been found that the individual organs of *G. elwesii* and *G. nivalis* produce alkaloids of different types. In the roots of *G. nivalis*, lycorine and tazetin-type alkaloids predominate, in the bulbs - tazetin-type, in the leaves - lycorine-type, and in the flowers - haemanthamine-type. In the roots, bulbs and leaves of *G. elwesii* - alkaloids of the homolicorin type were found, and in the flowers - of the tyramine type (publ. B1).

- The geographical distribution of the different chemotypes of *G. elwesii* and *G. nivalis* has been established. The populations of *G. elwesii* form one large and compact group and another small group of 2 populations containing lycorine-type alkaloids. The populations of *G. nivalis* form a western and eastern group, according to their geographical location in Bulgaria (publ. B2).

- Different alkaloid biosynthetic pathways have been determined in *G. nivalis* and *G. elwesii* from sympatric populations. Alkaloids formed by ortho-para'oxygen coupling of O-methylnorbelladin and tyramine-type alkaloids dominate in *G. elwesii*. Alkaloids formed by parapara'oxygen coupling of O-methylnorbelladin dominate in *G. nivalis*. The differences in the secondary metabolism of the two species are a good chemotaxonomic basis for proving the discreteness of the two species (publ. B1).

- It has been proven that the alkaloid composition of *G. elwesii* and *G. nivalis* remains constant over the years and under plant cultivation conditions. These results support the thesis that alkaloids in the genus *Galanthus* are genetically determined and can be used as chemotaxonomic markers (publ. B2).

- In total 29 alkaloids have been identified in alkaloid fractions of *Narcissus* cv. *Hawera*, 13 of which are of the *Sceletium* type. Of these, 2-oxo-mesembrine and 2-oxo-epi-mesembrenol are new alkaloids, and lycorine is the main alkaloid in the total alkaloid extract (publ. G14).

- GC-MS of silvlated samples has been found to be a suitable approach for the analysis of alkaloids in *Narcissus* cv. *Hawera* (publ. G14).

- Sceletium type alkaloids have been identified for the first time in in vitro grown plants of Narcissus pallidulus and Narcissus cv. Hawera (publ. G15).

- Differences in the metabolic profile of diploid and autotertaploid *H. papilio* plants have been found and over 60 primary and secondary metabolites have been identified (publ. G18).

- It has been found that autotetraploidization in *H. papilio* (Ravenna) Van Scheepen plants leads to an increase in the content of galantamine and hemantamine in the leaves of the plant, compared to diploid plants (publ. G18).

- Differences in the metabolic profile of *H. papilio* in different organs and parts of plant organs have been found. Thus, alkaloids and sugars are in higher concentration in the underground organs of *H. papilio*, while phenols, phytosterols, organic and fatty acids are in higher amounts in the leaves (publ. G21)

- The scientific information has been summarized and it has been found that gas chromatography and mass spectral analysis (GC-MS) is a suitable method for metabolic profiling

of plant samples from in vitro cultures. The methods for sample preparation and the methods for analyzing the results of GC-MS analysis are presented in a synthesized form (publ. G23).

- The content of three steroidal saponins and rutin was determined in 16 populations of *Tribulus terrestris* L. Significant quantitative differences in the main compounds between the populations were observed and the deposits that are promising for obtaining raw materials for the pharmaceutical industry were determined (publ. G9).

The other important direction in the scientific research of Assist. Prof. Dr Boryana Sidzhimova is the study of the biological activity of various plant extracts. The more significant contributions in this direction are:

- Strong acetylcholinesterase inhibitory activity was established for alkaloid extracts from *G. elwesii* and *G. nivalis* (publ. B7), as well as for essential oils of some representatives of the Asteraceae and Lamiaceae families (publ. G16).

- Very good inhibitory activity of alkaloid extracts from *G. elwesii* and *G. nivalis* on seed germination and root growth of *Lolium perenne* was established. These results also have a scientific and applied nature, as they can serve as a basis for the development of new biocidal agents against pests and weeds (publ. B7).

- It was found that there is no direct correlation between the strong inhibitory activity of acetylcholinesterase and the inhibition of seed germination (publ. B7).

- It has been proven that bioactive fractions of Amaryllidaceae alkaloids exhibit growth inhibitory activity against endophytic fungi isolated from the same plant (publ. G20).

- For the first time, it has been found that the alkaloid extract of the aboveground mass of *Narcissus cv. Hawera* can damage the DNA of *Chlamydomonas reinhardtii* (publ. G22).

As more significant contributions in the direction of "botanical research and taxonomic decisions" can be mentioned:

- the collection, systematization and analysis of the available information on the genus *Galanthus* in Bulgaria, with a revision of all herbarium specimens (publ. B4).

- the establishment of great variability in the studied morphological characters at the intraspecific and interspecific level (publ. B3). For example, the anatomical structure of the leaves in *G. elwesii* and *G. gracilis* is similar, while that of *G. nivalis* is different, while the embryological characteristics of *G. elwesii*, *G. nivalis* and the plants identified as *G. gracilis* are the same (publ. B3).

- the establishment that *G. elwesii* and the plants identified as *G. gracilis* have the same surface characteristics, including epicuticular waxes based on the SEM analysis of pollen and seeds (publ. B3).

The most important contributions in the areas of "plant cultivation" and "distribution and monitoring", which have not only scientific, but also scientific-applied character, are:

- the successful adaptation of in vitro plants of *H. papilio* in soil cultivation conditions. The effective method for propagation of selected genotypes together with adequate techniques for cultivation and obtaining biomass can establish *H. papilio* as a new and competitive source for industrial production of galantamine (publ. B5).

- the greater biomass of diploid plants of *H. papilio* in the first year of their development in hydroponic cultures compared to autotetraploid plants (publ. G18).

- the horological analysis of the modern distribution of the species *G. nivalis* and *G. elwesii* on the territory of the country - a particularly valuable piece of information missing so far. It has been established that *G. elwesii* is widespread in Bulgaria, while *G. nivalis* has a limited distribution. Sympatric populations of both species have also been identified (publ. B4).

- the determination of the ecological characteristics and monitoring of the populations of the species of the genus *Galanthus* in Bulgaria, which are included in the National Biodiversity Monitoring System (publ. B4).

- the resource assessment of 32 *Tribulus terrestris* L. localities in Northern Bulgaria and the identification of 19 of them as economically important for the pharmaceutical industry for the production of Tribestan (publ. G8).

- the determination of the distribution, area, condition and resource potential of 10 selected populations of *Colchicum autumnale* L. (publ. G10).

- the identification of a high degree of vulnerability of *Rhodiola rosea* L. to anthropogenic influence and climate change (drought, reduced snowfall) in the localities of the species in the Rila Mountains (publ. G13).

5. Assessment of the candidate's personal contribution

The candidate's contribution to the research conducted is undeniable, since in 5 of the presented scientific works, Assist. Prof. Dr. Boryana Sidzhimova is the first and/or corresponding author. The candidate in the competition has undoubtedly developed the necessary scientific competence and skills for identifying alkaloids of different skeletal types using gas chromatographic and mass spectral methods, for analyzing and summarizing the results obtained, for resolving taxonomic problems through the application of anatomical, comparative-embryological, genetic and morphological analyses, for research on biological activity, for the cultivation of promising species, as well as for the ecological features and monitoring of plant species. This acquired competence is a continuation, expansion and upgrading of the research from the dissertation work on species of the genus *Galanthus*, which over time have been expanded to other plant species. I believe that Dr. Boryana Sidzhimova, 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.

CONCLUSION

The documents and materials presented by Assist. Prof. Dr. Boryana Sidzhimova 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 Regulations for the Implementation of ZRASRB of the Bulgarian Academy of Sciences and the Regulations of IBER-BAS. The candidate in the competition has presented a significant number of scientific works published after the materials used in the defense of the PhD thesis. The candidate's works contain original scientific and applied contributions that have received international recognition, as a representative part of them have been published in journals and scientific collections published by international academic publishing houses. The scientific qualification of Assist. Prof. Dr. Boryana Sidzhimova is undoubted.

The results achieved by Assist. Prof. Dr. Boryana Sidjimova in scientific research activities fully comply with the specific requirements of the Regulations of IBER-BAS for the application of the ZRASRB.

After familiarizing myself with the materials and scientific works presented in the competition, analyzing their significance and the scientific, scientifically applied and applied contributions contained in them, I find it reasonable to give my positive assessment and to recommend to the Scientific Jury to prepare a report-proposal to the Scientific Council of IBER-BAS for the election of Assist. Prof. Dr. Boryana Sidjimova to the academic position of "Associate Professor" at IBER-BAS in the field of higher education: 4. "Natural Sciences, Mathematics and Informatics"; Professional field: 4.3. "Biological Sciences"; Scientific specialty: "Botany".

25.11.2024

Reviewer:

(Prof. Dr Antoaneta Trendafilova)