STATEMENT

Maria Prokopova Geneva – associate professor IPPG-BAS

For promotion procedure to the academic position "associate professor"
at the Institute of Organic Chemistry with Centre of Phytochemistry (IOCCP), BAS
in the professional field 4.2. Chemical science, scientific speciality “Bioorganic chemistry, chemistry of natural and physiologically active compounds”

For participation in the announced promotion procedure in the Newspaper of State 43/31.05.2019 and on the web site of IOCCP, BAS, for an associate professor for the needs of the Laboratory of Chemistry of Natural Substances has submitted documents only one candidate: assistant professor Kalina Moneva Danova, PhD, from IOCCP, BAS. Presented by assistant professor Danova set of materials fully meet the requirements of the LDASRB and the Regulations on the Conditions for Procedures for Acquisition of Academic Degrees and Occupation of Academic Positions at IOCCP, BAS for the promotion to the academic position "Associate Professor".

The scientific publications of assist. Prof. Danova, presented for participation in the promotion procedure is in the field of creation of a protocol for *in vitro* propagation of *Hippophae rhamnoides* L., *Hypericum* species, *Artemisia alba* Turra, tracing the influence of the used growth regulators on the qualitative and quantitative composition of essential oils and some physiological indicators. The purpose of the research is the controlled cultivation of medicinal and aromatic plants with defined properties. The applicant has applied 7 scientific publications in group B - publications, which are referenced and indexed in world-recognized databases of scientific information, in which the candidate is referred to as the author of correspondence and are equated to habilitation work. Of these, 2 publications have Q1, 1 with Q2, 4 with Q3 factors. In Group D, the candidate has applied 14 scientific publications in editions that have been referenced and indexed in world-famous databases of scientific information other than habilitation work. The distribution of scientific works according to the relevant Q factors is - 3 papers are with Q1, 4 with Q2, 1 with Q3 and 3 Q4, as well as 3 books (in which the candidate is an only author). Also presented are: a list of 112 citations in scientific publications, monographs and collective volumes, referenced and indexed in world-famous scientific information databases; a list and abstracts of the candidate's participation in 98 Scientific Forums; a list of her participation in 14 research projects. All this is a good indicator of the quality of the candidate's scientific output.

Assistant Professor Danova in 2000 graduated from the Faculty of Pharmacy, Medical University, Sofia with a PhD in Phytochemistry. In 2010 she successfully defended his PhD thesis in the field of Plant Biotechnology and Plant Physiology at the Sofia University “St. Kl. Ohridski". In 2010 he started working as a chemist at IOCCP, BAS since 2001 is an assistant professor.

I think that the scientific interests of assistant professor Kalina Danova, are dedicated to a very topical and important field. Her scientific research is focused on the selection of type and concentration of growth regulators added to the nutrient medium to obtain the optimal protocol for in vitro propagation of the investigated medicinal and aromatic plants - *Hippophae rhamnoides* L., some plants from Genus Hypericum, *Artemisia alba* Turra and influence on the qualitative and quantitative composition phytochemicals (essential oils and phenolic compounds) as well as on some physiological parameters.

Indisputable success of assist. Prof. Danova's is her many years of efforts to create and characterize the protocol for *in vitro* propagation of *Artemisia alba* Turra. In her studies, she found that the morphological development of plants due to the addition of growth regulators (auxin - 0.5 and 1.0 mg L^{-1} indole-3-butyric acid (IBA) and cytokinin 0.2 mg L^{-1} 6-benzyladenine (BA)) in the MS culture medium affects the terpenoid biosynthetic pathway, the concentration of polyphenols and flavonoids, and photosynthetic parameters. Addition alone of IBA provokes the development of the root system over the shoot, associated with increased biosynthesis of oxidized monoterpenes, decreased concentration of polyphenols and flavonoids until the thylakoid macro organization is not changed. The combined addition of IBA and BA leads to a lack of root formation, callus formation and increased sesquiterpenes in essential oils, a decrease in the content of bioactive forms of endogenous cytokinins; disturbance of the structural organization of the photosynthetic apparatus and the architecture of chloroplasts, increase in the content of phenols and flavonoids. By identifying sesquiterpenes, it has established the presence of different sesquiterpenes in the aboveground and underground parts of the plants.

In the aboveground parts of *A. alba*, for the first time, it has identified ten new sesquiterpene alcohols of which seven germacrenes, a eudesmane, a guaiane and an oplopane, as well as the phenolic compounds hispidulin, jaceosidin, desmethoxycentaureidin, and dicaffeoyl esters of quinic acid. It has been found that the flowers are rich in common phenols and flavonoids and has high antioxidant activity. In plant-samples collected from the Balkan Peninsula, flavones and flavonols are found, unlike those in the Mediterranean countries that contain only flavonols.

The candidate has conducted valuable studies on *Hippophae rhamnoides* L. (Blind) flavonoid content. The determined abundance of flavonoids found in the leaves of the *H. rhamnoides* (the higher values in the samples collected from the natural habitat near Varna, compared to the sample from the Botanical Gardens collection), makes the plant with potentially high pharmacological value and favourable for its cultivation in Bulgaria. Another important contribution of the candidate is to determine the best nutrient medium for induction of axillary bud from *H. rhamnoides* L. - adding 0.2 and 0.3 mg L^{-1} BA in ¼ MS medium.

An important part of the research conducted by the candidate is devoted to the genus *Hypericum*. The relationship between antioxidant enzymatic and non-enzymatic activity and the polyphenolic content of the *in vitro* propagated hypericin not producing *H. calycinum* is investigated. The content of polyphenolic compounds in the hypericin non-producing *H. calycinum* was found to be higher than in the hypericin producing species *H. tetrapterum*, *H. rumeliacum* and *H. richeri*. In the evaluation of NO-antiradical activity in hypericin-producing species, lower activity in *H. tetrapterum* was measured compared to the other two species belonging to the evolutionarily more developed Drosocarpium section.

Assist. Prof. Danova compared the effect of vitamins in Gamborg and MS nutrient medium on the development, enzymatic and non-enzymatic antioxidant capacity of hypericin producing *H. rumeliacum* and *H. tetrapterum* and hypericin non-producing *H. calycinum*. It found that the Gamborg medium vitamins more increased the levels of hypericin, MDA and H_2O_2 in *H. rumeliacum* and *H. tetrapterum* compared to MS vitamins. She hypothesized that the increased content of hypericin and stress markers MDA and H_2O_2 may be related to the physiological stress that plants undergo during *in vitro* propagation. On the other hand, the high content of hypericin may cause additional oxidative stress. It implies that hypericin also plays a role in the adaptation of the plant to changes in the environment.

In conclusion, the assist. Prof. Danova's achievements are not only of fundamental importance but are also applicable to agricultural practice for the studied medicinal and aromatic plants cultivation. The results provide a solid basis for implementing a protocol with optimal concentration and combination of IBA and BA, resulting in both the production of plants with high photosynthetic efficiency and high accumulation of phenolic compounds and essential oils with a specific composition according to the requirements of the pharmaceutical industry.

The submitted documents for the competition have been prepared in a very precise manner and are designed in an understandable and easy to evaluate way. In the habilitation report, assist. Prof. Danova described in detail and correctly the complete scientific contributions, the experiments of which are carried out at IOCCP-BAS and are published in prestigious scientific journals with co-authors by scientists from other scientific institutions. However, it is not clear

from the habilitation Report what its personal involvement in: developing the overall concept; the team of specialists formation, proficient in methods in various fields; experimental design; processing, analyzing and interpreting results for writing articles or submitting a report for oral or posters presentation in scientific forums. This is also my only remark to assist. Prof. Danova.

CONCLUSION

The documents and materials presented by Assist. Prof. Kalina Danova fulfils all the requirements of the Law on the Development of the Academic Staff in the Republic of Bulgaria (LDASRB), the Regulations for the implementation of the LDASRB, the Regulations for the implementation of the LDASRB of the BAS and the Rules of the IOCCP-BAS. The candidate submitted a sufficient number of scientific papers, published after the materials used in the defence of the Doctor 's thesis. The applicant's works have original scientific and applied contributions that have received international recognition by being published in journals published by international academic publishers. The scientific qualification of the applicant is undoubtedly undeniable. Achieved by assist. Prof. Danova results in the research activity, fully completed the specific requirements of the IOCCP-BAS Regulations for the implementation of the LDASRB.

After getting acquainted with the materials and scientific works presented in the competition, analysis of their importance and the scientific, scientific-applied and applied contributions contained therein, I find it justifiable to give my positive assessment and to recommend to the Scientific Jury to prepare a report proposal to the Scientific Council of IOCCP-BAS for the selection of assist. Prof. Kalina Moneva Danova at the academic position of Associate Professor at IOCCP-BAS in the professional field 4.2. Chemical Sciences, scientific speciality "Bioorganic chemistry, chemistry of natural and physiologically active substances".

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(assoc. prof. M. Geneva)