

REFEREE REPORT

By Prof. Pavlina Aleksandrova Dolashka, D.Sc. from Institute of Organic Chemistry with Center for Phytochemistry – BAS,

in competition for awarding the academic position of "Professor" in the professional field 4.2 Chemical Sciences, specialty "Bioorganic chemistry, chemistry of natural and physiologically active substances" for the needs of the Laboratory of Biologically Active Substances (LBAS), Institute of Organic Chemistry with the Center of Phytochemistry – BAS, Plovdiv published in the State Gazette no. 43 of 31 May.2019

In the competition for "Professor" in the professional field 4.2 Chemical Sciences, scientific specialty "Bioorganic chemistry, chemistry of natural and physiologically active substances" for the needs of the laboratory of Biologically Active Substances, IOCCP - BAS participated only one candidate, Assoc. Prof. Petko Nedyalkov Denev.

1. General presentation of the received materials

The presented for reviewing materials completely correspond to the requirements of the Law for Scientific Development in Bulgaria, the Rules of its Application as well as of the Internal Rules and Regulations of the Institute Organic Chemistry with Center for Phytochemistry (IOCCP) for acquiring the academic position "Professor".

Pack of documents for reviewing: Completed table (Annex 1 of the IOSCF Rules) certifying meeting the requirements for occupying the academic position of "professor"; 2. Scientific autobiography; 3. Diploma of educational and scientific degree "doctor" (copy); 4. Diploma for the academic position "Associate Professor" (copy); 5. An abstract of the dissertation for the educational and scientific degree "Doctor"; 6. Extended habilitation reference for the scientific contributions of Bulgarian and English with a common list of publications; 7. List and copies of publications by Indicator B (equivalent number articles); 8. List and copies of publications and certificate of

registration utility model by indicator D; 9. List of participations in scientific events with attached evidence. 10. List of quotations not submitted in another competition; 11. List of participations in research projects with attached evidence material; 12. Additional information about the scientific activity - list and evidence material.

2. Biographical data

Assoc. Prof. Denev graduated from the University of Food Technology - Plovdiv in 2005 with MS in "Biotechnology". He continued his education in the same institute, where he acquired a PhD degree (2011) in the scientific specialty "Technology of biologically active substances". Dr. Denev is a works in the Institute of Organic Chemistry with Center for Phytochemistry - BAS, Laboratory of biologically active substances – Plovdiv (LBAS).

3. General characteristics of the applicant's activities

Evaluation of the applicant's scientific and applied scientific activity

Dr. Denev is an author of 88 scientific papers, of which 47 are in peer reviewed scientific journals with a total IF >100, cited 762 times. He is a co-author of 3 monograph chapters, 2 patents and utility models and participated in 19 scientific projects with national funding and 3 projects with the companies. In 2013 D-r Denev was awarded with the "Pythagoras" Grand and with Young Scientist Award "Prof. Marin Drinov".

Assessment of educational and pedagogical activity

Dr. Denev is a head of a scientific team on applied research in the field of biochemistry, food and pharmacy in the Institute of Organic Chemistry with Center for Phytochemistry - BAS, Laboratory of biologically active substances – Plovdiv (LBAS).

The scientific contributions

Central to the research of Laboratory of biologically active substances, over the last 15 years, are the fruits of aronia (*Aronia melanocarpa*). Black chokeberry (*Aronia melanocarpa*) fruits are also among the richest sources of polyphenols and anthocyanins in plant kingdom, and suitable raw material for production of functional foods. The scientific contributions of Dr. Denev's research work have both fundamental and applied character and are directed mainly to studying the chemical composition of medicinal and economically important plants and their biological activity.

The main scientific and applied contributions from presented 31 publications can be summarized in three thematic areas:

1. Investigation of chemical composition and biological activity of medicinal and economically important plants:
2. Utilization of waste from the essential oil industry to obtain new products;
3. Investigation of chemical composition and biological activity of pectic polysaccharides.

Scientific contributions to the area 1: Investigation of chemical composition and biological activity of medicinal and economically important plants.

The scientific contributions of area 1 include two subareas:

The first subarea : Investigation of chemical composition and biological activity of fruit and functional foods of aronia (*Aronia melanocarpa*);

Dr. Denev's group has studied on the chemical composition of 23 aronia fruit samples from two harvest years – 2016 and 2017. They demonstrated that chokeberry samples differ significantly in both the content and the composition of organic acids, sugars and phenolic compounds. The differences in the chemical composition of the fruits resulted in functional foods that differ significantly in their chemical composition and antioxidant activity. As the strongest antioxidants among aronia polyphenols the quercetin and epicatechin were found.

The black chokeberry proanthocyanidins are proven as the most potential antimicrobial agents in the fruit. The fruits are particularly rich in anthocyanins, which are very instable during processing and storage of aronia products. The tested compounds of different phenolic co-pigments and herbal extracts on color intensity of aronia anthocyanins provoked different co-pigmentation effect, manifested by hyperchromic and bathochromic shifts.

The use of selected herbal extracts as co-pigments by Dr. Denev opens realistic prospects for development of aronia functional foods with improved sensory properties and biological effects, due to enhanced color and anthocyanin stability.

Also the other interesting results on the effect of aronia juice on learning ability, memory and brain morphology of aged rats was presented. These results indicate that aronia juice induced ameliorating changes in the ability of old rats to learn tasks and improved their locomotor functions.

The second subarea: Investigation of chemical composition and biological activity of other medicinal and economically important plants.

The phytochemical composition and biological activity of Bulgarian fruit, vegetables, herbs and the health effects of flavonoids and their role in healthy nutrition are presented in this part of investigations of Dr. Denev.

A high antioxidant activity Blackberry (*Rubus fruticosus*) leaves, chokeberry (*Aronia melanocarpa*) leaves, hawthorn (*Crataegus monogyna*) leaves, lady's mantle (*Alchemilla glabra*) aerial parts, meadowsweet (*Filipendula ulmaria*) aerial parts and raspberry (*Rubus idaeus*) was identified. Also, the antioxidant activity of extracts from *Valeriana officinalis*, *Melissa officinalis*, *Crataegus monogyna*, *Hypericum perforatum*, *Serratula coronata* and their commercially available combinations Antistress 1 and Antistress 2, used as food supplements and recommended for chronic fatigue, are represented. With the most pronounced antioxidant activity are the extracts of *M. officinalis* and *H. perforatum*.

Also the antioxidant activities of various flavonoid glycosides isolated from dwarf elder (*Sambucus ebulus*), the antioxidant activity of *Salvia tomentosa* essential oil, the kinetic parameters of co-pigmentation between strawberry anthocyanins and caffeic acids were determined.

For the first time the chemical composition and antioxidant activity of the extremely rich in polyphenolic substances fruits of *Chaenomeles maulei* was investigated. The sour taste of the juice and its low pH were due to the high content of organic acids (malic, quinic, citric, shikimic, ascorbic, oxalic). The carbohydrates were presented by glucose, fructose, galactose, xylose, rhamnase and arabinose.

The chemical composition, nutritional value, amino acid and fatty acid profiles of several plants as a dried stevia (*Stevia rebaudiana*) leaves, considered as one of the strongest natural sweeteners, and quinoa (*Chenopodium quinoa*) are represented.

Scientific contributions to the area 2: Utilization of waste from the essential oil industry to obtain new products

The subject of this area is a chemical characterization of extracts derived from waste biomass from essential oil industry. They have found that lavender (*Lavandula angustifolia*) and melissa (*Melissa Officinalis*) waste, are rich on polyphenols (especially rosmarinic acid) and aroma compounds, and exhibited high antioxidant and antimicrobial activity. The effect of lavender and melissa waste is to shelf the life of bread.

The synthesis of nanoparticles mediated by *Rosa damascena* waste extracts is also presented and their application for an electrochemical sensing of hydrogen peroxide and vanillin.

Scientific contributions to the area 3: Investigation of chemical composition and biological activity of pectic polysaccharides

In the frame of this area, Dr. Denev analysed for the first time the polysaccharide composition in 11 European and in particular Bulgarian medicinal plants with high *in vitro* complement-fixation activity and high *ex vivo* intestinal immunomodulatory effect. The pectin-type acid heteropolysaccharides in Bulgarian medicinal plants with a pronounced immunomodulating activity with immunostimulating and anti-inflammatory potential have been proven.

Implementation activities

The results obtained by Dr. Denev are of practical application and can serve as a basis for the development of value-added by-products from cultivated Bulgarian herbs.

4. Assessment of the applicant's personal contribution

Although many of the works of Dr. Denev have been published in a collaboration with other authors, his role is obvious.

5. Critical comments and recommendations

I have no critical comments on the materials presented and the research. I am highly recommending Dr. Denev to continue his studies on the structure and biological effects of bioactive compounds and mainly on studying the chemical composition and biological activity of medicinal and economically important plants and the waste from their processing, for the development of nutraceuticals and functional foods.

6. Personal impressions

I know Assoc. Prof. Denev and my personal impressions are that he is dedicated to his work as a scientist, able-bodied and purposeful, which has undoubtedly led to significant achievements, both scientific and practical.

CONCLUSION

The presented results, shows that Assoc. Prof. Petko Nedyalkov Denev has in-depth theoretical knowledge and professional skills in the scientific specialty "Bioorganic

Chemistry, Chemistry of Natural and Physiologically Active Substances", demonstrating qualities and skills for conducting research with obtaining original and significant scientific contributions.

The research has been carried out using a modern methodology and instrumentation. The obtained results are published in high ranked peer reviewed journals in the fields of chemistry and biochemistry, and contribute a lot to basic science and practice. The papers are cited hundreds of times in the world literature. Two patents are also issued. Dr. Denev fully satisfies the requirements of the Law for Scientific Development in the Republic of Bulgaria, the Rules of its Application as well as of the Internal Rules and Regulations of the Institute Organic Chemistry with Center for Phytochemistry for acquiring the academic position of "Professor".

Because of that I am giving my positive assessment of his research, obtained results and scientific contributions and I propose to the Honorable Scientific Jury to award the academic position of "Professor" of Assoc. Prof. Petko Nedyalkov Denev in the field of higher education: 4. "Natural Sciences, Mathematics and Informatics"; professional field: 4.2. "Chemical Sciences"; scientific specialty "Organic Chemistry, Chemistry of Natural and Physiologically Active Products".

10 September 2019

Sofia

Reviewer:

(Prof. Pavlina Dolashka, DSc)