OPINION

By Dr Valentina Todorova Toneva,

Professor at the University of Plovdiv

Of a dissertation to acquire the educational and scientific degree "Doctor".

Field of higher education: 4. Natural sciences, mathematics and informatics.

Professional direction: 4.3. Biological Sciences.

Doctoral program: Botany.

Author: Donika Petrova Gyuzeleva

Topic: Biological and phytochemical study of plants from the Bulgarian flora with potential for biotechnological application.

Scientific supervisors:

Prof. Dr. Plamen Stefanov Stoyanov, University of Plovdiv (UoP).

Prof. Dr. Anelia Veselinova Bivolarska, Medical University of Plovdiv

1. General description of the presented materials

By an order No. PD-21-2058 of 11/15/2024. of the Rector of UoP, I have been appointed as a member of the scientific jury to ensure a procedure for the defence of a dissertation on the topic "Biological and phytochemical study of plants from the Bulgarian flora with potential for biotechnological application" for the acquisition of the educational and scientific degree "doctor" in the field of higher education 4. Natural sciences, mathematics and informatics, professional action 4.3. Biological Sciences, Doctoral Program - Botany.

The dissertation author is Donika Petrova Gyuzeleva, a full-time doctoral student at the Department of Botany and Biological Education, with scientific supervisors Prof. Dr. Plamen Stefanov Stoyanov and Prof. DSc. Anelia Veselinova Bivolarska – both from UoP.

The set of materials presented by Donika Petrova Gyuzeleva is by Article 36 (1) of the Regulations for the Development of the Academic Staff of the UoP and includes the following documents:

- 1. A request to the Rector of the PU to disclose the procedure for defending a dissertation work.
- 2. Resume in European format.
- 3. Minutes from the departmental council related to reporting the readiness to open the procedure and preliminary discussion of the dissertation work.

- 4. Dissertation work.
- 5. Abstract.
- 6. List of scientific publications on the topic of the dissertation.
- 7. Copies of scientific publications.
- 8. Declaration of originality and authenticity of the attached documents.
- 9. Certificate of compliance with the minimum national requirements.
- 10. Set of documents 1-9 on electronic media.
- 11. Brief biographical data of the doctoral candidate

Donika Gyuzeleva obtained a bachelor's degree in medical biology and a master's in microbiological control and food safety at Up. She is the deputy head of the "Human Resources" Department at Up and an assistant at the Department of Botany and Biological Education at the UoP.

2. Relevance of the topic

The problem developed in the dissertation work is undoubtedly relevant in scientific and scientific-applied terms. In general, it refers to the multifaceted study of several (medicinal) plants containing more than usual amounts of secondary metabolites such as flavonoids, glycosides and lipids that could be used medicinally to treat important social diseases. On the other hand, similar plants can fall into technological processes of important productions that supply essential food for humans. This increases the relevance of the problem and the specific tasks developed in the dissertation.

3. Knowing the problem

The dissertation's high-quality and detailed literature review shows excellent knowledge of the problem and creative handling of the collected scientific literature, which also applies to the methodological part.

4. Research methodology

To achieve the set goal and obtain an adequate answer to the tasks solved in the dissertation, suitable modern research methods, the latest and highly sensitive modern equipment, and innovative approaches were chosen. All this allows for high reliability of the obtained results, which is essential to reach a higher level of discussion, considering that the plant species used have been widely studied in the past and now.

5. Characterization and evaluation of the dissertation work and contributions

The dissertation follows the classic scheme of construction, starting with a good introduction to the problem, setting the primary goal of the work and the specific research tasks, the latter of which can be said to be highly diverse - on the border of reasonable diversity for a dissertation like the one under consideration. In this regard, the bulky anatomical part outweighs the exposition of the primary purpose. Very well with skill and knowledge of the problem, the main research methods are selected in the "Materials and Methods" section, significantly analytical and preparative chromatographic and mass spectrometric metabolomic approaches.

The central part of the dissertation, "Results and discussions," is formally and logically divided according to the data for the two studied plant groups—*Marrubium* and *Centaurea*. This also enables the grouping of the type of research and the obtained results. The discussion was conducted with high expertise and based on reliable data, which allowed us to derive certain regularities in the presented conclusions.

The dissertation analyses scientific and scientific-applied achievements in two main directions: detailed and precise botanical and anatomical characteristics of the plants used, which have a more classical form and content. These characteristics more precisely confirm earlier studies and could be the basis for further taxonomic and metabolomic studies with significant innovation and scientific-applied contributions.

Of the contributions, the data on the rich metabolic composition with numerous secondary metabolites and microelements of the two species of *Marrubium*, which could have a medical application, are of particular importance. Their phytochemical composition confirms their diverse bioactive effects, making them valuable sources for biotechnological productions. Here, the antitumor effects should be more carefully commented on since the data rest exclusively on in vitro cell studies that provide giant exposure concentrations to the isolated cells and tissues.

Undoubtedly, however, these lines of research in the dissertation would have applications in the pharmaceutical field and are significant contributions of significance and development perspective to science and practice.

6. Evaluation of the publications and personal contribution of the doctoral student

Four publications are presented that reflect the results of the dissertation, all of which are scientific articles in specialised publications. The publications are classic scientific publications presenting research results on the studied plant species. The editions used are of different importance, with three belonging to Q1 and one to Q4 (in a national edition without an impact factor). Three publications have a relatively high impact factor; two are open-access publications of the MDPI editions (Molecules and IJMS). This number and quality of publications can be considered a high achievement for the dissertation and represents the work well done for the scientific community, evidenced by the already established seven citations (Scopus). However, the publications are only from the last 2 years.

The doctoral student is the first author of three publications and the last author of one. He is not a corresponding author for any publications.

I believe that, to a large extent, the publications, work performed, and conclusions and contributions made are the personal work of the doctoral student. In the metabolomic studies, substantial help was also received from the co-authors. No plagiarism was found during the dissertation review.

7. Abstract

The abstract is made according to the requirements of the relevant regulations and reflects the main results achieved in the dissertation.

8. Recommendations for future use of dissertation contributions and results

The dissertation could benefit from a greater concentration on biochemical and metabolomic studies, careful commenting on antitumor effects, and more generalisable conclusions.

The thesis contains many biochemical and metabolomic studies that are worth pursuing at a more detailed level to identify specific molecules with future medical and nutritional applications.

CONCLUSION

The dissertation contains scientific, scientific-applied, and applied results that represent an original contribution to science and 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, and the relevant Regulations of UoP.

The dissertation shows that doctoral student Donika Petrova Gyuzeleva possesses in-depth theoretical knowledge and professional skills in the scientific speciality of botany. It also demonstrates qualities and skills for independent scientific research.

Due to the above, I confidently give my positive assessment of the conducted research, presented in the dissertation work, abstract achieved results and contributions, and I propose to the honourable scientific jury to award the educational and scientific degree "Doctor" to Donika Petrova Gyuzeleva in the field of higher education: 4. Natural sciences, mathematics and informatics, professional direction 4.3. Biological Sciences, Doctoral Program "Botany".

16/12/2024

Prepared the opinion:

Prof. Dr. Valentina Todorova Toneva