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R E V I E W

By **Professor Dr of Science. Nidal Tabit Shaban**, retired from the Forestry University-Sofia with the scientific direction "Plant Breeding-Vegetable Production" of a dissertation for awarding the educational and scientific degree "doctor" in: 4. Natural sciences, mathematics and informatics professional direction 4.3. Biological sciences, scientific specialty-Botany with code 01.06.11, Author – **Master Boryanka Dimitrova Traikova.**

Topic of the dissertation: **Hydroponic technologies as a means of protection and cultivation of medicinal and conservation-important plant species.**

Research supervisor: Prof. Dr. Marina Stanilova

Scientific consultancy: Assoc. Dr. Ina Aneva

General characteristics of the dissertation - volume and structure

By order No. 62 / 27.09.2024 of the Director of IBEI-BAN: I have been appointed as a member of the scientific jury to ensure a procedure for the defense of a dissertation work on the topic "Hydroponic technologies as a means of protection and cultivation of medicinal and conservation significant plants species." for the acquisition of the educational and scientific degree 'doctor' in the field of higher education 4. natural sciences, mathematics and informatics, professional direction 4.3. biological sciences, science specialty Botany with code 06.01.11. The author of the dissertation is Boryanka Dimitrova Traikova - a doctoral student of independent researcher with scientific supervisor Prof. Dr. Marina Stanilova and Scientific consultancy: Assoc. Dr. Ina Aneva. The set of paper materials presented by the doctoral student is in accordance with Art. 39 (1) of the Regulations for the conditions and procedures for acquiring scientific degrees and for occupying academic positions at IBEI-BAS.

Presented scientific work written on 176 pages. The structure meets the requirements of the normative documents with introduction, purpose and tasks, literature review, materials, method results and discussion. According to Article 9 of the Regulations on the conditions and procedure for acquiring scientific degrees and for holding academic positions in the Institute for Biodiversity and Ecosystem Research at the BAS, the chapters "Materials and methods, Results and discussion" are replaced by copies of the publications on the topic of the dissertation; Conclusion, Main conclusions, Cited literature. The list of cited literature includes 138 sources, 129 in Latin and 9 in Cyrillic. The discussion topic formed in this way

is suitable for scientific direction 6.1, plant breeding and the scientific specialty vegetable production.

Brief biographical data of the PhD student

The PhD student is well prepared in Russian and English, acquired during her studies. She acquired considerable knowledge and skills in working with a computer and modern scientific equipment during her regular studies at the Faculty of Agronomy of LTU-Sofia, under the master's program " Plant protection", as well as within the work and in the RGRR department at IBEI-BAS.

1. Significance of the researched problem in scientific and scientific-applied terms.

Preservation of the biodiversity of plant species used for human food after drastic climate changes is in the focus of scientific circles all over the planet. Adaptation of new methods in their reproduction and their introduction into agriculture is the focus of attention of the author of the proposed dissertation work and her scientific supervisors. The presented work is a continuation of their efforts to achieve the best results in this direction.

The studied plant species, characterized by valuable medicinal and aromatic properties, have a resource deficit and potential for cultivation. The species are from the Bulgarian flora, often endemic to Bulgaria or the Balkan Peninsula. In accordance with normative documents, such as the Law on Biological Diversity (2002), the Law on Medicinal Plants (2000), the Law on Protected Areas (1998), the Red Book of the Republic of Bulgaria (1984, 2015), as well as the one received from Bulgarian producers, the author and its scientific supervisors selected the studied plant species. In a number of genebanks in the world, propagation and cultivation of rare species applied as in geoponic, hydroponic, aeroponic and other methods, and invitro, invivo, etc. methods used for storage.

Until now, attempts at hydroponic cultivation made with some species characterized by valuable medicinal and aromatic properties, with a resource deficit and potential for cultivation. A positive evaluation deserves the proposed reference to the literary sources, which covers the most recent and significant publications on the problem, which have been critically read, highlighting the controversial and controversial points of view. The aim and tasks clearly and precisely formulated, and follow logically from the literature review. The material and methods correctly described. The results presented accurately, without any errors and discrepancies between the text and the data contained in the figures and tables. The

discussion of the results is thorough, and shows the doctoral student's free handling of the literary material. The conclusions are adequate to the obtained results. Contributions correctly worded.

2. Precisely formulated objectives and tasks of the dissertation.

The stated purpose of proposals for reviewing scientific work precisely formulated. In the long term, the main goal is to stabilize the natural populations of plant species with conservation status or to create pilot plantations of medicinal and aromatic plants. To achieve them, a number of specific tasks have been set, namely: creating protocols for propagation and accelerating the growth of selected species of medicinal plants and species of conservation importance, characterized by difficulties in propagation (low germination and/or slow growth), by means of the application of various hydroponic technologies to eliminate the negative effect of climate change. The comparative evaluation of the used methods is influenced by the plant species, the composition of the nutrient solutions, and the chemical composition of the substrates and the physiological reaction of the studied plants. In the course of the implementation, it planned to establish opportunities for the hydroponically propagated individuals to retain the biosynthetic ability characteristic of the species, as well as with regard to secondary metabolites. The set goals are related both to the clarification of purely scientific questions and to the possibility of practical application of the obtained results. The developed scientific product acquires scientific importance as well as scientifically applicable value, especially when scaling the implementation of the created protocols and adapting the methods used. In the dissertation, the applied scientific articles are in the field of biodiversity conservation, the applied methods are a co-existing factor ensuring the achievement of the set goal.

3. Evidence that the dissertation work developed independently and does not literally repeat the topic and a significant part of the content of the work presented for the acquisition of the educational and scientific degree "doctor".

Suggestions for reviewing a dissertation is the personal work of the doctoral student. The development appears as a continuation of the efforts of its scientific leaders in protecting the biodiversity of plant species used for human food after the drastic climate changes. The successful realization of the set task is also due to their joint efforts in equipping phytotrons with different types of hydroponic systems, differing in the way of supplying the nutrient solution, and suitable for cultivating plants from vegetative organs (cuttings, leaves) or for

germinating seeds contribute to the successful implementation of this kind of research, especially species with a conservation status or those that synthesize and accumulate biologically active substances in their roots. The choice of a suitable substrate, or lack thereof, depending on the requirements of the plant species, is also important. The application of hydroponic cultivation enters some medicinal plants with difficult reproduction. One of the main advantages of hydroponics related to accelerated growth. In this regard, it is appropriate to apply it, especially in slow-growing plants, under other methods and conditions. The hydroponic and aeroponic systems, allowing the testing of alternative biotechnological methods for seed germination and vegetative propagation of plants from the target groups of species. The dissertation presented to us meets the requirements of the IBEI regulations.

To achieve the set goal, the following tasks are correctly and consistently set:

1) Investigation of the possibilities for vegetative reproduction from leaves and

Cuttings of target species by testing different hydroponic systems (Haberlea rhodopensis, Vaccinium vitis-idaea, Arctostaphylos uva-ursi, Thymus longedentatus, T.pannonicus, T. zygioides).

2) Comparative analysis of growth acceleration of bulbs/ Lilium rhodopaeum and Hippeastrum papilio/, obtained in vitro or from seeds, when testing different hydroponic systems and substrates.

3) Study of the possibilities for seed propagation of medicinal and

Conservation important plant species (Haberlea rhodopensis, Alkanna tinctoria, Salvia officinalis and Echinacea purpurea) by testing different hydroponic systems and substrates.

4) Increase survival rate and growth efficiency by hydroponic cultivation of ponies (Alkanna tinctoria).

5) Adaptation of the hydroponically cultivated plants to soil substrate in a phytotron room and subsequent acclimatization to greenhouse conditions and outdoors in the ex situ collections.

6) Comparative analysis of the results of chromatographic determination of the biologically active substances of hydroponically grown plants compared to the control ones (Salvia officinalis, Hippeastrum papilio, Thymus longedentatus, T. zygioides).

The significant part of the work developed to realize the set goal, namely acquisition of the educational and scientific degree "Doctor".

4. Degree of knowledge of the state of the problem and correspondence of the used literature.

The dissertation student knows very well the state of the problem and creatively evaluates the literary material, which allows her to formulate the purpose and tasks of the dissertation work and discuss the obtained results in depth. The rich set of literary sources indicated in the review includes 138 sources, 129 in Latin and 9 in Cyrillic. The literature review examines hydroponic technology in a detailed and exceptionally scientific style in a comprehensive overview. In 22 pages, the author describes the history and development of hydroponics, the role of technology in agriculture, classification of hydroponic methods - water or substrate, according to the environment in which the root system develops, the types of substrates - inorganic, organic and soilless mixtures, the requirements for the nutrient solution and its adaptation to the type and conditions of cultivation, physiological equilibrium of nutrient solutions, environmental conditions in closed rooms (lighting, temperature, humidity, carbon dioxide CO_2), determining the need for nutritional elements by the appearance of plants, advantages and disadvantages of technology compared to the conventional method of growing plants. And last but not least, he skilfully presented the methods of aeroponics, a classification of hydroponic methods according to the method of feeding the nutrient solution. In this reference, the author shows significant knowledge in this direction, familiar with the local literature dealing with this subject, as well as the world literature fund.

In the attached scientific publications, the author showed enviable knowledge in the field of biodiversity conservation of plant species used for human food after drastic climate changes. The cited literature covers the subject of the dissertation work. The cited authors correctly cited.

5. Availability of a justified and developed theoretical model of the study.

As a continuation of the accumulated data from long-term research conducted at IBEI, the selection of plant species in the dissertation work are economically and socially. Thanks to the active participation of the doctoral student in many projects dealing with this direction of her research work and helped in the correct selection of significant plant species: medicinal and aromatic plants with market demand, most with resource deficits, as well as species of conservation importance. Thus, the research corresponded with the institute's mission, in which the main areas of work in the RGRR department, as all target species studied in projects developed by the RGRR department, in which the doctoral student took part. In addition, the selection of the plant species, the subject of the dissertation, made in a way that allowed the

study of the main methods of reproduction: vegetative (through leaves, cuttings, and bulbs) and seed (through seeds and pods), with the application of appropriate hydroponic methods.

6. Correspondence of the chosen methodology and research methodology with the set goal and tasks of the dissertation work.

The chosen methods for the fulfilment of the tasks of the dissertation work are in full compliance. The skilful combination in such an interdisciplinary dissertation work, with the endless support of its scientific supervisors, contributed positively to the correct definition of the goals and tasks and the stages for their successful implementation. Another important merit of the dissertation work is the complexity of the research approaches and the wide spectrum of parameters determined during the research work. The presented scientific publications and the successful participation in international forums for the popularization of the obtained results represent confidence in the methods used for the reliability of the specified results, on which the contributions of the dissertation work based.

7. Availability of own contribution in the collection and analysis of empirical data.

The dissertation shows that the candidate has in-depth theoretical knowledge of the relevant specialty and abilities for independent scientific research. The work contains theoretical summaries and solutions to scientific or applied scientific problems that represent an original contribution to science. Because of the conducted research, valuable scientific contributions obtained, some of which are of an original nature, namely:

1) New protocols created for the propagation of medicinal and conservation significant plant species with the application of hydroponic technologies at the laboratory level through: vegetative propagation (Haberlea rhodopensis, Thymus longedentatus, T. pannonicus, T. zygioides and Vaccinium vitis-idaea) and seed propagation (Echinacea purpurea, Salvia officinalis, Alkanna tinctoria).

2) Improved the protocol for accelerated in vitro propagation of slow-growing bulbous species (Lilium rhodopaeum and Hippeastrum papilio) by upgrading with a new step: applying hydroponic cultivation of the in vitro obtained bulbs to accelerate their growth.

3) Haberlea rhodopensis plants obtained for the first time by hydroponic propagation, using the leaf, vertical Green Diamond (best results) and horizontal Aeroflo-20 aero-hydroponic systems.

4) Plants of three types of thyme (Thymus pannonicus, T. zygioides and the Balkan endemic T. longedentatus) obtained by vegetative propagation from cuttings, using hydroponic technologies.

It is essential to point out that the results obtained in the dissertation represent a basis and perspective for future scientific studies, for solving controversial issues and clarifying new aspects of the researched issues. The author of the presented dissertation took an active personal part in the planning and execution of the assigned tasks, in writing and shaping the work, as well as in shaping the contributions and conclusions of this author's work.

8. Evaluation of publications on the dissertation work

The dissertation includes eight publications reflecting the experimental results obtained during the development of the dissertation work. Personal involvement of the doctoral student The dissertation and the contributions to it are largely the personal work of the doctoral student; the fact that she is first author on 7 of the articles supports this claim.

9. Abstract

The abstract was prepared according the requirements and regulations of IBEI and correctly reflects the main results obtained in the dissertation.

10. Critical remarks and recommendations

Some critical comments and recommendations can be made to the dissertation work: - **Literature review** - As a note to the literature review, it is obvious that in many places there are no citations and it is not clear who is the source of the information. At the end of this section, no conclusion justifies the need to conduct such an interdisciplinary scientific study due to the lack of such developments. This omission does not reduce the quality of the proposed section and confirms the author's literary awareness and theoretical training.

Contributions - These are very descriptive and largely repeat the results. They can be shorter and clearer. It is good to divide them as original or of a confirmatory nature. They can be scientific, applied science or applied in nature. The critical remarks and recommendations made do not detract from the dissertation work, and are only intended to serve the future work of the dissertation doctoral student.

11. Personal impressions

I have very positive personal impressions of the doctoral student, received during the lecture courses "vegetable production and greenhouse vegetable production" conducted by me within the AF Bachelor program at LTU-Sofia. A positive impression also left by her performance with the mentioned publications and participation in scientific projects.

12. Recommendations for future use of dissertation contributions and results

I recommend that in her future research, the doctoral candidate should focus on a more detailed characterization of the obtained results and activation of international cooperation for the improvement of this kind of developments, combining abiotic and biotic factors, as well as in familiarization with new other systems.

CONCLUSION

The dissertation contains scientific, scientific-applied and applied results, which 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, the Regulations for the Implementation of IBEI.

The presented materials and dissertation results fully correspond to the specific requirements of IBEI. The dissertation work shows that the doctoral student Boryana Dimitrova Traikova possesses in-depth theoretical knowledge and professional skills in the scientific specialty - Botany with the code 01.06.11, demonstrating qualities and skills for independently conducting scientific research.

Due to the above, I confidently give my positive assessment of the conducted research, presented by the above-reviewed dissertation work, abstract, achieved results and contributions, and I propose to the honourable scientific jury to award the educational and **scientific degree "doctor" to Boryana Dimitrova Traikova in the field of higher education in natural sciences, mathematics and informatics, professional direction 4.3. Biological sciences, science speciality Botany with code 06.01.11.**

25.10.2024 **Reviewer:** **Prof. Dr. of Science Nidal Tabit Shaban**