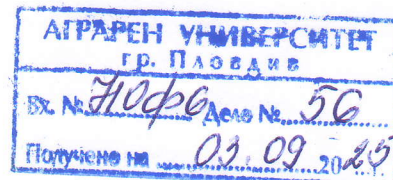


OPINION



on the dissertation for the degree of "Doctor of Science"
in the professional field 4.3. Biological Sciences, scientific specialty Genetics

Author of the dissertation: Prof. Dr. Bojin Maximov Bojinov from the Agricultural University – Plovdiv

Topic of the dissertation: Use of molecular markers in genetic diversity studies and for DNA profiling

Member of the scientific jury: Prof. Dr. Andon Vassilev Andonov from the Agricultural University - Plovdiv, professional field 4.3. Biological Sciences, scientific specialty Plant Physiology, appointed as a member of the scientific jury by Orders No. RD 16-753 / 12.06.2025 and RD 16-801/ 24.06.2025 of the Rector of the Agricultural University - Plovdiv.

1. Relevance of the scientific problem considered in the dissertation

The dissertation thesis of Prof. Dr. Bojin Bojinov examines the problem of the use of molecular markers for the analysis of genetic diversity and for DNA profiling in various organisms (plants, animals, pathogens). The prepared literature survey objectively and critically presents the available information on the problem. It becomes clear that the problem is not new and the opportunities that the use of molecular markers provides for accelerating the selection process in plants and the selection and breeding activity in animals, the registration and control of seed and planting material and of reproductive material in animals, are well known, but still insufficiently used in our country. The reasons for this state of affairs are different, but some of them are related to a lack of information about the advantages and disadvantages of the marker systems used depending on the goals and specific objects of the study, material and technical problems, the lack of standardization of protocols, etc. In this aspect, the relevance of the problem in the developed dissertation thesis is high, and the research conducted is well-reasoned.

2. Purpose, hypotheses, tasks and research methods

The purpose of the developed dissertation thesis is clearly formulated - to select a molecular marker system for identifying genetic diversity in different types of organisms (plant, animal and microbial), which will ensure reliability and reproducibility of the results and possibilities for use in different directions, namely -

for breeding purposes and for control of variety testing and seed production / breeding of local breeds.

The working hypothesis of the dissertation is that from the many known marker systems, one can be selected that most fully meets the set goal. To achieve the goal, 3 main tasks and 4 subtasks have been selected, which are correctly formulated and logically interconnected.

Molecular marker systems were used, including both dominant (AFLP, ISSR, CAPS) and co-dominant (SSR, RFLP) analyses on a large number of work objects: plants - cotton, tomatoes, tobacco, pepper, wheat, corn, barley, paulownia, animals - autochthonous goat breeds and fungi - pathogens of the genera *Phytophthora* and *Fusarium*.

3. Visualization and presentation of the obtained results

The dissertation is structured traditionally – Introduction, Literature review, Aim and objectives, Material and methods, Results, Discussion, Conclusions, Contributions and References. It is written on 256 pages, of which the sections Results and Discussion occupy 50%. The cited references are 280. The 17 photos presented illustrate some of the experimental objects used in the study. The main results are presented in 63 figures and 33 tables, which with minor exceptions (a few figures) are clear and sufficiently informative. The results are subjected to statistical analyses with appropriate programs and are considered separately for each of the experimental objects used.

4. Discussion of the results

The discussion of the obtained results is conducted thoroughly and competently. It clearly demonstrates the rich scientific information of the dissertationist on the studied problem, as well as his practical experience in the use of molecular markers in highly phylogenetically different objects (plants, animals, eukaryotic microorganisms). The results of the application of different molecular systems to specific work objects are critically analyzed and their advantages and disadvantages are revealed depending on the purpose of the study. The use of the ISSR marker system for identifying genetic diversity is justified due to its ability to identify polymorphism in varieties, lines and breeds, the relatively easy methodology, the good reproducibility of the results, as well as its possible use, in addition to selection purposes, for RHS testing.

5. Contributions of the dissertation work

As a result of the research conducted by Prof. Bojinov, significant scientific and applied scientific contributions of an original nature have been obtained. Due to the limited scope of the opinion, I will only mention some of them:

1. Original scientific contribution with the characterized ability of a set of dominant (i.e. AFLP, ISSR, CAPS) and co-dominant (RFLP, SSR) molecular marker systems for the detection of genetic diversity in plant and animal organisms of local origin.

2. Original scientific contribution is the proven ability of the ISSR marker system to identify intraspecific diversity in phytophthora fungi.

3. Original scientific and applied contribution is the information on new marker/trait associations for a number of economically valuable traits in local varieties of cotton and tomatoes.

4. An original scientific and applied contribution is the mapping of a significant number of molecular markers in cotton, enriching the existing genetic map with loci with significant effects in local agro-ecological conditions.

6. Critical comments and questions

I have no critical comments.

7. Documents and scientometric data related to the dissertation work

The documents and materials submitted by Prof. Bojinov in connection with the procedure for defending a dissertation work for the acquisition of the scientific degree of "Doctor of Sciences" fully meet 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 Law in the Republic of Bulgaria (RILRB) and the Regulations for the Implementation of the Law at the Agricultural University – Plovdiv (RILAU; specifically Art. 59, item 3).

Prof. Bojinov has presented 19 scientific publications, related to the dissertation thesis, and 4 patents for created cotton varieties. The scientometric assessment of the indicated publications and patents, according to the the Regulations for the Implementation of the Law in the Republic of Bulgaria, amounts to 254 points with a minimum required standard of 100. Nine of the submitted publications received a total of 167 citations (152 from the list of publications and 15 in citing publications), which form 334 points with a minimum requirement of 100.

8. Conclusion

The dissertation of Prof. Dr. Bojin Bojinov is an in-depth study on the use of molecular markers for the study of genetic diversity and for DNA profiling in various biological organisms. It uses modern molecular marker systems (AFLP, ISSR, CAPS, SSR and RFLP analyses) and statistical methods and has obtained significant scientific results, representing an original contribution to science. The dissertation meets all the requirements of the LDASRB, RILRB and RILAU for the acquisition of the scientific

degree "Doctor of Sciences". It shows that Prof. Bojinov possesses in-depth theoretical knowledge and professional skills in the field of genetics and molecular biology.

Based on the above, I confidently give my positive assessment of the conducted research, the achieved results and contributions, and I propose to the esteemed scientific jury to award the scientific degree "Doctor of Sciences" to Prof. Dr. Bojin Maximov Bojinov in professional field 4.3. Biological Sciences, scientific specialty "Genetics".

08/15/2025

Prepared by:

Prof. Dr. Andon Vassilev

