

PhD Thesis Acceptance Report
Research Discipline Council of Biological Sciences
Jagiellonian University in Kraków

Candidate's name and surname: Mohamadreza Mirzaei

PhD Thesis Title: Chemical defense systems in *Arabidopsis thaliana* leaves

Thesis Supervisor: dr hab. Ewa Dubas

Assistant Supervisor / Second Supervisor/ Co-supervisor (if applicable): dr. Kenji Yamada, prof. UJ

Reviewer: dr hab. inż. Ewa Grzebelus, prof. URK

1. The topic of the dissertation

In a natural ecosystem, the pathogen-plant-insect relationship has various consequences for each other. Pathogens and insects feed on plant tissues, which in most cases leads to plant damage. Therefore plant species, have developed specialized defense systems both to protect themselves and to reduce damage. Different types of plant pathogens and herbivores are fought by means of various changes in plants, such as changes in gene expression, detoxification of glucosinolate metabolism, signal transduction, cell wall modification, and Ca²⁺ dependent signaling. These complex defense mechanisms can take place in specialized cells such as myrosin cells and ER body-containing cells as observed in plants of the *Brassicaceae* family. Further research on plant defense systems, which is the subject of the doctoral dissertation presented here, is an extremely important part of ongoing research in the biological sciences. The results can have fundamental meaning for ecology, biotechnology and agriculture. In particular, a better understanding of these mechanisms can contribute to the development of new crop varieties with increased resistance to pathogens and pests.

2. The candidate's knowledge

Section 'Introduction' of presented dissertation demonstrates general theoretical knowledge of the Applicant in the discipline of the biological sciences. In this section, the Author presents the most important information on: (1) chemical defense system in *Brassicaceae* plants, (2) single and dual-cell type chemical defense strategy, (3) significance and characteristic of ER bodies in plant defense systems, and (4) the role of glucosinolates, jasmonic acid and auxins in plant defense systems and the main factors of their regulation. The text is coherent, well-organized and has been prepared on the basis of numerous (nearly 70 items), including the most recent (2020-2024) literature published in recognized scientific journals in the biological sciences. In addition, for better understanding of the content, it contains 9 figures, including 7 by the Author. In my opinion, the content presented in this section is appropriately selected and comprehensive (although perhaps too concise in places) in the context of the dissertation topic. This demonstrates the Author's good orientation to the current state of knowledge in the research topic undertaken.

3. Independence of the candidate

The Author has clearly formulated general and specific research objectives. He selected appropriate plant materials (wild type of *At* and a large collection of appropriately selected mutants obtained from various mutant collections and research units) and appropriate research methods.

The scope of research activities included cross-pollination of selected accessions, molecular analyses of nucleic acids and proteins, and microscopic analyses including using confocal microscopy imaging. The set of research methods used is broad and properly selected to meet the research objectives. The text of the Materials and methods section provides an initial overview of the range of methods used sufficient to understand the course of the experiments conducted and the analyses performed.

The number as well as the value of the results obtained is impressive. They are documented in 25, mostly very elaborate but readable, figures. The results were logically presented, synthetically but clearly discussed, and correctly interpreted in relation to the available literature. At the same time, the author presented interesting interpretations of the identified findings.

In conclusion, all the above-mentioned arguments strongly demonstrate the ability of the Author of the presented dissertation to carry out scientific work independently.

4. Originality of the dissertation

The doctoral dissertation submitted for evaluation by Mr. Mohamadreza Mirzaei is a valuable scientific study providing an original solution to the scientific problem on chemical defense systems in plants, in particular on mechanisms of hormonal regulation of the development and functionality of myrosin cells and ER body-containing cells. The results suggest that long-term MeJA treatment induced the expression of defense-related myrosinase genes (*TGG1* and *TGG2*) independent of the canonical JA signaling pathway and highlight the importance of the MeJA treatment in gene expression changes. On the other hand it was shown, that in the single-cell type chemical defense strategy, position of cells plays a crucial role in the differentiation of ER body-containing cells. JA and auxin are the potential signaling molecules responsible for the ER body-containing cell differentiation influencing on production of ER bodies and regulating vascular distribution pattern, respectively. The results presented here point to complex interactions between MeJA and IAA, myrosinase cells, ER body-containing cells and vascular tissue, which may coordinate effective plant defense responses to pathogens and insects. In the era of new genomic techniques, these results may be of particular importance for developing new source materials for breeding programs aimed at production crop varieties with increased resistance to biotic factors.

5. Questions and/or criticisms to which the Reviewer expects the candidate to respond during the defence

Some comments and questions:

- In the subsection 'Plant materials', a listing of accessions used in a table with name, origin and phenotype of mutants would be helpful in quickly identifying the materials analyzed.
- What was the aim of cross-pollination conducted? Seed propagation of valuable accession or an analysis of the heritability of certain genes – if so, which ones?
- What was the biological replication in all experiments? A single plant or a mixed sample – if so, from how many plants?
- What is the mechanism of cell wall staining using PI?
- Considering the results obtained, would it be possible to edit the genome in terms of increasing the resistance of plants to biotic factors? What crop would the Author choose for such research and what genome editing scheme would he propose?

Other minor comments to the text not required to respond during the defense:

- some of the reference sources pointed in the text were not listed in the literature list, e.g.: Xue et al. 1995; Yamada et al. 2011; Lu et al. 1996;
- fig. 9: in the legend to the figure abbrs. for MYC2 and JAR1 were not explained;
- table 12-13: the final volume of the solutions has been omitted;
- fig. 17: abbrs. for CBB is not explained.

I, hereby, declare that the reviewed PhD thesis by **Mohamadreza Mirzaei** meets the criteria pursuant to art. 187 of Act of 20 July 2018 The Law on Higher Education and Science (Journal of Laws of 2018, item 1668, as amended) and request that the Research Discipline Council of Biological Sciences of the Jagiellonian University in Kraków accepts **Mohamadreza Mirzaei** for further stages of doctoral proceedings in the field of exact and biological sciences, in the discipline of biological sciences

YES/~~NO~~

I, hereby, request that the thesis is accepted with distinctions

~~YES~~/NO

Ja, niżej podpisana stwierdzam, że recenzowana rozprawa doktorska Pana **mgr. Mohamadreza Mirzaei** spełnia warunki określone w art. 187 Ustawy z dnia 20 lipca 2018 r. Prawo o szkolnictwie wyższym i nauce (Dz. U. z 2018 r. poz. 1668 z późn. zm.) i wnioskuję do Rady Dyscypliny Nauki biologiczne Uniwersytetu Jagiellońskiego w Krakowie o dopuszczenie Pana **Mohamadreza Mirzaei** do dalszych etapów postępowania ws. nadania stopnia doktora w dziedzinie nauk ścisłych i przyrodniczych w dyscyplinie nauki biologiczne.

TAK/~~NIE~~

Ja, niżej podpisany wnioskuję o wyróżnienie rozprawy doktorskiej.

~~TAK~~/NIE



Signed by /
Podpisano przez:

Ewa Marta
Grzebelus

Date / Data: 2025-
06-16 08:14

Kraków, June 16, 2025

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date

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Reviewer's signature