

## OPINION

Лесотехнически университет  
Агрономически факултет  
№ АФ-1097#3  
СОФИЯ 12.12.2022

on a Dissertation for obtaining an educational and scientific degree „Doctor“ in: Field of higher education 6. Agricultural sciences and veterinary medicine, Professional field 6.2 Plant Protection, scientific specialty “Plant Protection (Phytopathology)”

**Author of the PhD thesis:** Rosan Said Shaalan is a part time PhD student at the Department of Plant Protection, Faculty of Agronomy, University of Forestry, Sofia.

**PhD thesis title:** Impact of *Beauveria bassiana* and *Metarhizium anisopliae* on the interactions between *Cucumis sativus* L., cotton aphid (*Aphis gossypii* Glover) and Cucumber mosaic virus (CMV)

**Member of the Scientific Jury: Prof. DSc. Rossitza Borissova Batchvarova**, field of higher education 6. Agricultural sciences and veterinary medicine, professional field 6.2 Plant protection, scientific specialty scientific specialty “Plant Protection (Phytopathology, Virology, Herbology etc.)”, appointed a member of the scientific jury by order № ЗПЦ- 642 / 05.12.2022 by the Rector of the University of Forestry, Sofia.

### 1. Relevance of the problem

Cucumber is a vegetable crop, the economic importance of which varies according to the region of the world. It is most popular in Europe, the USA and Asia.

The popularity of its fruit is on its essential nutrients (e.g., vitamins and minerals) and other beneficial substances (e.g., soluble fibers and antioxidants). The cucumber is one of the most important greenhouse crops, its intensive production is more depended to the rapid spread of insects such as cotton aphid, *Aphis gossypii* Glover. The damages caused by it reflect on production, and also by its ability to transmit plant viruses such as cucumber mosaic virus (CMV).

CMV is one of the most common and serious plant viruses worldwide, infecting over 1200 plant species and being spread by more than 80 aphid species. Furthermore, CMV cannot be controlled by chemical pesticides and therefore its control depends on the eradication of its vectors including *A. gossypii*.

*A. gossypii* is mainly controlled by chemical insecticides for a long time and resulted in the development of resistance by the pest. These problems have brought about the need to introduce other sustainable, effective and safe alternative biological control strategies against *A. gossypii*. Over the years, various biological control tactics have been evaluated in many parts of the world for the management of this pest.

Entomopathogenic fungi (EPF), *Beauveria bassiana* (Balsamo) Vuillemin and *Metarhizium anisopliae* (Metchnikov) Sorokin (Ascomycota: Hypocreales) have also been shown to be promising alternatives in the bio agriculture. They are currently being developed, registered, marketed and used as biopesticides against many insect species including aphids.

All this determines the relevance and significance of the topic developed in the dissertation.

### 2. Degree of knowledge of the state of the problem and creative interpretation of the literature review.

In the dissertation work PhD student presented a perfect review of the literature on the problem including more than 400 scientific publications. She is citing very correctly and discussing her results related with the results achieved of scientist working in this area.

### **3. Purpose, tasks, hypotheses and research methods. Correspondence of the chosen research methodology with the set goal and tasks of the dissertation work.**

The main goal of this research is to study entomopathogenic endophytes as potential alternatives to chemical insecticides against *Aphis gossypii* on cucumber plants and their possible use to control this pest and subsequent transmission of many destructive viruses like Cucumber mosaic virus (CMV). The current project aims to study also metabolomics of CMV infected cucumber plants to find out the effects of applied endophytes *Beauveria bassiana* and *Metarhizium anisopliae* (EPF) on the induced defense mechanism against the virus.

The purpose of the dissertation is correctly formulated, and 8 main tasks are precisely and clearly stated, the implementation of which has led to the results.

The doctoral student has successfully mastered the necessary for the purpose of the dissertation modern research methods as: methods used in phytopathology, virology, entomology, physiological, biochemical, molecular methods for identification of microorganisms and aphids, Scanning Electron Microscope (SEM), etc.

### **4. Visualization and presentation of the obtained results**

The presented dissertation is a purposeful research work that complements and expands the knowledge of the problems. It is formed according to the classical model of 160 pages adopted in the Republic of Bulgaria, as the illustration and presentation of the results are achieved with 7 tables, 44 figures and 13 appendices.

The structure of the dissertation is well balanced, as the literature review is 35 pages, materials and methods - 16 pages, results and discussion - 37 pages and conclusions and contributions - 2 pages.

### **5. Discussion of the results and the used literature.**

The obtained results are summarized and interpreted correctly, in a good scientific style. In discussing them, the sequence of the presented problem in the literature review is followed, which gives value and clarity to the exposition. All experiments were performed strictly, which makes the results reliable. The discussion on them shows an in-depth knowledge of the issue and 416 literature sources were used.

### **6. Contributions of the dissertation work.**

From the developed dissertation work, the PhD student made 7 significant scientific contributions and 4 applied contributions as a results of the research work.

The main achievements are related with colonization, penetration and the effect on plant growth of entomopathogenic endophytes *B. bassiana* and *M. anisopliae* on cucumber. It was found that cucumber plant colonization by fungal entomopathogens is generally negative to aphids *A. gossypii*, reducing their population size.

One important result of the current research is that cucumber plants, treated with endophytes showed strong metabolic response to CMV infection with highest percentage of metabolites in amino acid derivatives.

This method of applying endophytes to plants could increase the resistance of cucumber varieties to CMV infection.

The applied contribution of the PhD thesis will be very useful for the farmers involved with bio production of vegetables.

#### **7. Assessment of the degree of personal participation of the PhD student in the contributions**

The dissertation's results, presented contributions and conclusions drawn are entirely the result of the student's work.

#### **8. Critical notes and questions.**

I have no critical remarks or questions to the doctoral student.

#### **9. Published articles and citations.**

In connection with the dissertation, the doctoral student has published 2 scientific articles- in Journal of Plant Protection Research and in Horticulturae. Also one paper was published in the Book of Proceedings of the IX International Scientific Agriculture Symposium AGROSYM 2018", Bosnia and Herzegovina.

She also has 16.7 and 11.1% participation in two other scientific publications that are not related to the dissertation. In total, the doctoral student has 57.33 points.

Three citations of the presented publications are attached..

#### **10. Assessment of the publications based on dissertation work: number, nature of the editions in which they are published. Reflections in science- use and citation by other authors**

The PhD student has presented two papers, based on the results of dissertation. One of them is published and the second accepted for publishing in refereed and indexed edition in world- renowned scientific information databases (WoS and Scopus) journals as Journal of Plant Protection Research and Horticulturae. In all papers she is a leading author with 70% contribution.

She has two oral presentations (Agrosym 2018, 04 – 07 October 2018, Bosnia and Herzegovina; Sixth CRSL Research Conference– American University of Beirut AUB, 2022) and one poster presentation (Conference at Balamand University, Lebanon (2018) at international scientific forums.

The presented abstract reflects objectively the structure and content of the dissertation work.

#### **CONCLUSION:**

Based on the learned and applied by the doctoral student, different research methods, correctly performed experiments, summaries and conclusions, I believe that the presented dissertation meets the requirements of ZRASRB and the Rules of the Forestry University for its application, which gives me the reason to evaluate it **POSITIVE**.

I allow myself to suggest to the esteemed Scientific Jury that it also vote positively and to award **Rosan Said Shaalan** the educational and scientific degree "Doctor" in the field of higher education 6. Agricultural sciences and veterinary medicine, professional field 6.2 Plant Protection, scientific specialty "Plant Protection (Phytopathology)".

Date: 09.12.2022

**OPINION PREPARED BY:**

(Prof. DSc. Rossitza Batchvarova)