Reviewer’s Report on the PhD thesis

Reviewer: Kiril Bahcevandziev


Supervisor: Professor RNDr. Ladislav Havel

Study programm: PSP Botany (Field of Study: PFS Forest Phytology)

The thesis consists of 125 pages of the text (including figures, tables and reference lists).

This thesis presents a study related with the establishing of a test system for investigation of the ability of two conifer trees Abies alba and Picea abies to undergo somatic embryogenesis under abiotic heavy metal (Cd\(^{2+}\) and Pb\(^{2+}\)) stress. As a part of a study Ing Biljana Đjordjević analysed how different concentrations of Cd\(^{2+}\) and Pb\(^{2+}\) can affect the cell viability in early somatic embryos of A. alba and P. abies.

The topic of the thesis is actual, important and highly relevant especially because it combines abiotic stress with plant biotechnology/biochemistry and statistical methods taking care of the environment. There are very few works related with this subject that gives the thesis highest source and importance for the future works.

The Abstract is concise, well written and clearly shows the work developed.

Suggested observations:

It is not need to use abreviations (such as ESE, ATP and ROS) in this part. If the reason was to save the number of characters, there are other ways to do it.

The keywords are acceptable, maybe “abiotic stress” can be used instead of “phytochelatins”.

List of abbreviations is well done.

Suggested observations:

Some adjustments should be done: in the list ESE is before EDTA but shuld be oposite, first EDTA, ET, ETC and ESE ....); also in case of PCS and PCs should be oposite, PCs and after PCS.

Introduction is short, clear and very good written.
Question 1. The candidate mentioned Cd\(^{2+}\) and Pb\(^{2+}\) as soil pollutants. Is there any data that can show their presence in Czech soils?

Question 2. Why did the candidate use, in the work, slow growing conifer plants as potential phytoremediators? What percentage of the Czech forests do they occupy? Can suggest any important faster growing tree in Czech Republic, that can be used as phytoremediator?

Aim of the research is concise and clear.

Suggested observations:

In the text there is ESEs as abbreviation. Should be, when used for the first time in the main text, in an extensive form (early somatic embryos) and after that use ESE between brackets.

The Literature review is succinct, covering the main relevant theories and well structured. It reflects the work idea.

Suggested observations:

The figure legend is written in the same way (letter type, size and distance between lines) as the main text. Should be with smaller letters and lower distance between lines.

Legend of some figures (when there are two secondary figures not marked) should call attention to these figures and be marked (A and B or left and right), in the text.

Page 31: first sentence – “In this thesis effect of most important phytohormones for somatic embryogenesis i.e. auxins, cytokinins and ABA will be discussed” - should be changed as there is no such discussion in the thesis.

Page 38: should exist a connection between chapter 3.9 and both sub-chapters 3.9.1. and 3.9.2. Missing a sentence in the text of chapter 3.9 (or last paragraph) that can link the other two sub-chapters.

The Material and selected methods of study are adequate with respect to the aim of the thesis.

The Results chapter is well organized and follows the previous descriptions as in the Literature Review as in the Material and methods chapters. Tables and Figures are mentioned in the text in adequate way as it is required in it.

Suggested observation:

The figure and table legends should be adapted as mentioned above (see Literature Review analyses).
Discussion is well-argued, summarising the main contributions for the research done, showing interesting and thoughtful reflections on the research process and orientations for future work in the studied field.

Suggested observations:

In some sentences there is “etc.” on the end. It should be omitted. This abbreviation “etc.” can mean a lot of things.

Page 84: First paragraph, in the sentence: “in case of P. abies Pb²⁺ treatment, unexpectedly already lowest concentration had highest …… in the heavy metal stress defense” is missing something.

Page 86: In the sentence (line 7): “From our results can be seen that in Picea abies SOD was approximately 4-fold higher than in Abies alba.” Needs to explain when it happened? With both or in a case of one of heavy metals?

The final chapter, Conclusions, summarizes Cd²⁺ and Pb²⁺ tolerance in coniferous species and their future possibilities as phytoremediators. The problem is challenging.

I can conclude that the thesis is well-written, with very few clerical errors, appreciate style and good layout. The candidate demonstrates creative capacities in her research field and the thesis meets the required standard of a doctoral thesis to justify the award of a PhD.

Coimbra, 10 October 2017

Kiril Bahcevandziev, Professor Dr.