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Referee report on Ph.D. Thesis of Mgr. Adéla Hýlová entitled “Study of molecular mechanism and biological activity of strigolactones”

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The submitted Ph.D. thesis is devoted to the developing and optimizing bioassays that describe the activity of strigolactones, plant hormones, but also other biostimulants, such as polyamines. The topic of dissertation is timely. There are more than 200 studies, refereeing to strigolactones and their biological activities, published in last 18 months.

Formally, thesis is well structured into chapters and contains all usual items like the lists of references, tables, figures and abbreviations used in the text. The first five chapters contain the comprehensive literature review on chemistry, biosynthesis, and signal perceptions of strigolactones, parasitic plants related to strigolactones, as well as bioassays for testing various functions and activities of strigolactones. The experimental and results are summarized in seventh and eighth chapters, while ninth and tenth chapters were dedicated to conclusions and reference list, respectively. The figures, tables, references and the text itself are processed according to standards. The bibliography of the work contains over 150 references.

Main results of the thesis, which are summarized in the eighth chapter, represent an original contribution of the author to the bioassaying of strigolactone mimics, as well as naturally occurring compounds with biostimulant characteristics. The parasitic weed germination assay, as well as a high-throughput MTT based germination assay has been optimized and set for routine testing. In addition, the MTT assay was validated and used for examination of biostimulant activities on *Arabidopsis* grown under salt stress conditions. These outcomes are significant for the researches working in the fields of plant biochemistry and physiology.

The results of the thesis have been already published in four papers, in highly impacted journals, which is strong evidence for the quality and applicability of author's research.

I am not a native English speaker; therefore I cannot give a proper evaluation of the language of the thesis. It is, however, obvious for a non-English reader that the thesis is written very carefully, in a clear and easily understandable way.

To conclude, author has clearly proven her ability to solve complicated scientific problems and to clearly present valuable data. There is no doubt about her potential both to perform an independent creative scientific work and to co-operate effectively in scientific teams.

I recommend this thesis to be accepted by the committee and I do believe that after successful defense Adéla Hýlová will be awarded the PhD scientific degree.

Questions and remarks:

1. The fact that phosphate deficiency enhances strigolactone biosynthesis has been mentioned several times through the thesis. Is it possible that presence of the phosphate influences their stability?
2. In order to provide better seed distribution for their pipetting in wells the MMT germination assay has been improved by addition of sterile agarose to a final concentration of 0.05 %. Are some other compounds/concentrations tested?
3. Stability of the new strigolactone mimics under different pH has been tested, and it was found that an oxo-IAA derivate is quite stable under pH above 7, which is good feature for field applications. Is there any information about germination rates of parasitic seeds under different pH conditions, especially pH above 7?
4. Why 0.1 % acetone was used as negative probe in germination assays, and DMSO for stability assay?



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