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## ОТЗЫВ

члена диссертационного совета на диссертацию Кузнецовой Ксении Андреевны на тему: «Система WOX-CLAVATA и ее мишени в регуляции развития запасяющего корня и спонтанных опухолей у редиса посевного (*Raphanus sativus* L.) / WOX-CLAVATA system and its targets in the regulation of storage root and spontaneous tumour development in radish (*Raphanus sativus* L.)», представленную на соискание ученой степени кандидата наук по научной специальности 1.5.7. Генетика

The thesis consists of three publication quality chapters as well as discussion section. The thesis is arranged well and written with clarity. I found no deficiencies and have no requests for corrections.

The thesis addresses an important topic of storage root development and mechanisms of spontaneous tumour generation in plants using radish as an economically relevant model system. Specific molecular genetics aim of this work builds on the knowledge about peptide control of plant development. The candidate addresses the mechanisms for the WOX4 and WOX14 transcription factors activating molecular pathways in and the CLE41 peptide functioning in root development of radish. The work elegantly utilizes radish lines from the genetic collection created and maintained at the Department of Genetics and Biotechnology of St. Petersburg State University to provide both genomic and genetic characterization of spontaneous tumor formation and root development.

In the first chapter, the candidate provides sequencing and analysis of the genomes of two lines of radish genetic with contrasting phenotypes in spontaneous tumour formation. The candidate uses state of the art next generation sequencing technologies to generate genomic data and applies standard analyses workflows for discovery of sub-set of genetic differences. As there are many genetic differences genome-wide between plant lines, the candidate designed prioritization of candidate variants based on pre-existing information about types of genes known to be involved in the pathways, showing knowledge of the field and relevant literature.

In the second chapter, the thesis provides a gene expression analysis of roots with RsCLE41 overexpression and studies the effect of RsWOX4 and RsWOX14 transcription factor overexpression on radish root development. Transcriptome analyses are carried out using standard analyses in the field and identify a total of 62 genes differentially expressed in radish roots with overexpression of

RsCLE41-1 compared to roots with overexpression of a negative control gene. The author classifies differentially expressed pathways, which provides new hypotheses about the role of abiotic and defense related stress response genes in storage root development as well as involvement of chromatin modifications. The study also identifies hormone response factors and transcription factors that can be amenable to storage root improvement either by breeding or bioengineering.

In the third chapter, candidate performs a bioinformatic genome-wide search for targets of RsWOX4 and RsWOX14 transcription factors. Computational analyses are followed up by molecular characterization of the interaction between the RsWOX4 transcription factor with the promoters of candidate genes using a yeast one-hybrid assay.

Overall, the methods are appropriate and well mastered, the data is critically evaluated, and the conclusions are solid. I have only minor concern as some of the protocols, such as high molecular weight DNA extraction are not publicly available, and some unpublished scripts are mentioned but not deposited. However, this should be evaluated in the context of research group rules and data management plan of the institute and funding agencies. The computational approaches are very well documented at the high standard for reproducibility. Molecular biology details related to gene amplification and quantification as well as data analyses are also well documented.

The scientific findings are critically evaluated and presented well with detailed and careful visualization. The literature review introduction not only provides review on the important topic but also generates ideas for the theoretical advancement of the field. The discussion and conclusions are appropriate and present original ideas.

The thesis provides sufficiently novel and significant information to be accepted. The author is first to perform sequencing of the lines from the collection and especially of lines with contrasting tumor phenotypes. Spontaneous tumors are rare yet are of high importance to biotechnology and understanding plant regeneration, therefore, insights from the presented work are significant for a wide audience. The genomic information adds not only to the specific field of study, but also to the comparative genomics projects and understanding biodiversity in general. Transcriptomic analyses present a well prioritized list of information for follow up studies. Finally, presented identification of the RsLOG3 gene encoding an enzyme of cytokinin biosynthesis is significant for improving our understanding of the factors regulating storage root development.

The main results of the thesis work have been presented at 6 international conferences and published as peer-reviewed articles in journals such as *International Journal of Molecular Science*, *Ecological Genetics*, *Plants*, and *Russian Journal of Plant Physiology* with candidate as 1<sup>st</sup> author with significant involvement in five articles, including obtaining plant material, experimental methodology, bioinformatics analysis, data visualisation, data validation, preparation of figures, writing most of the article, preparation of the article for publication. This demonstrates the impact of the work at both national and international levels.

I am happy to wholeheartedly recommend this thesis for acceptance.

С учетом всего вышесказанного полагаю:

Содержание диссертации Кузнецовой Ксении Андреевны на тему: «Система WOX-CLAVATA и ее мишени в регуляции развития запасящего корня и спонтанных опухолей у редиса посевного (*Raphanus sativus* L.) / WOX-CLAVATA system and its targets in the regulation of storage root and

spontaneous tumour development in radish (*Raphanus sativus* L.)» соответствует специальности 1.5.7. Генетика;

Диссертация является научно-квалификационной работой, в которой содержится решение научной задачи, имеющей значение для развития соответствующей отрасли знаний, либо изложены новые научно обоснованные технические, технологические или иные решения и разработки, имеющие существенное значение для развития страны

Нарушений пунктов 9, 11 Порядка присуждения Санкт-Петербургским государственным университетом ученой степени кандидата наук, ученой степени доктора наук соискателем ученой степени мною не установлено

Диссертация соответствует критериям, которым должны отвечать диссертации на соискание ученой степени кандидата наук, установленным приказом от 19.11.2021 № 11181/1 «О порядке присуждения ученых степеней в Санкт-Петербургском государственном университете» и рекомендована к защите в СПбГУ.

Член диссертационного совета

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