Evaluation of the diploma thesis of Alžběta Poštulková by her supervisor Magdalena Bohutínská

Alžběta's diploma thesis is a rare example of successfully linking genomic identification of a candidate repeatedly adaptive gene with the possible causes for its selection and its phenotypic consequences. Such a comprehensive connection is rare in evolutionary biology because it demands a thorough understanding and analysis of the model system at multiple levels: from evolutionary and population genetics, through ecological and organismal perspectives, to biochemical insights. This multidisciplinary approach requires a well-rounded expertise that is exceptionally rare for a diploma thesis.

In her thesis, Alžběta specifically demonstrated that the *FAR5* gene shows strong and repeated signs of selection in alpine ecotypes of *Arabidopsis arenosa* across five different mountain ranges. She further revealed that the repeatedly selected alpine allele alters the amino acid composition of this enzyme, impacting its substrate specificity and the composition of fatty alcohols in suberin. This finding adds the FAR5 gene to the list of 'evolutionarily significant' genes identified across various organisms, helping us understand the factors that make a gene adaptively relevant.

The thesis is written in an excellent English and is logically structured. The results are presented in three main parts. The first part discusses selection signals in the genomes of alpine lineages of *Arabidopsis arenosa*, with a focus on the FAR5 gene. The second part shows that the alpine and lowland alleles of the FAR5 enzyme change its substrate specificity, resulting in a detectable metabolomic phenotype. The third part tests which environmental conditions might have led to this change, suggesting that spring precipitation has the most significant influence on the spread of the alpine allele. These conditions also lead to the differentiation of several other genes, pointing to the possible role of the alpine FAR5 allele in regulating seed dormancy.

Throughout her study, Alžběta demonstrated remarkable scientific versatility and the ability to quickly master a wide range of methods and approaches. These included fieldwork in challenging alpine environments, population genomic analyses, advanced R programming, ecological analysis, climate data analysis, and molecular genetic and biochemical analyses in the laboratory. I dare to say that few master students are prepared for doctoral studies with such a rich skill set.

Alžběta also showed great independence in scientific management and communication. She completed much of her work during an independent internship at the University of Innsbruck, presented her experimental design and results at seminars in three international institutions (Max Planck Institute for Plant Breeding Research in Cologne, Institute of Ecology and Evolution at the University of Bern, and the Department of Botany at the University of Innsbruck), and at three international conferences. She funded a significant part of her research through her GAUK project and a mobility funds. Additionally, she supervised two high school students in their specialized research projects.

Overall, I find Alžběta’s contribution to her diploma project exceptional and propose evaluating her thesis, "The role of the *FAR5* gene in alpine adaptation of *Arabidopsis arenosa*," as excellent.

Finally, I would like to ask Bety two questions:

1. If you could change something at the beginning of the project or propose a different experimental approach, what would it be?

2. Among all the potential functional consequences of the shorter fatty acid alcohols in the alpine environment, which one would you choose for further study? Please suggest an experiment or series of experiments to test this.

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