

# Abstract

The repeated adaptation presents a unique opportunity to study the mechanisms of evolution in natural replicates. Repeated adaptation of *Arabidopsis arenosa*, a wild relative of the model organism *Arabidopsis thaliana*, to the alpine environment was previously studied. Genomic analysis by selection scans revealed a set of candidate alleles. Here, I present a functional follow-up study of a candidate alpine-adaptive allele of *FAR5*. Specifically, I asked: What are the characteristics of the alpine (*A*) allele compared to the foothill (*F*) allele? Is there any phenotypic effect of the two different alleles of *FAR5*? What are the environmental factors shaping the distribution of the alpine allele of *FAR5* among the populations of *A. arenosa*?

I first demonstrated that natural variation in *FAR5* played a role in adaptation to the alpine environment: I identified four SNPs that were positively selected in the alpine populations across all five high altitude colonizations of *A. arenosa*. Three of these SNPs are coding and are linked together, forming a distinct alpine allele of the *FAR5* protein. Using a unique crossing design based on a natural standing variation of the two identified *FAR5* alleles, I prepared carriers of the non-native alleles on the genomic background of foothill and alpine populations. I then conducted a transplant experiment into the alpine environment of Austrian Alps and examined the phenotypic effect of the two alleles. Due to the reported function of *FAR5* in wax biosynthesis in *A. thaliana*, I used a GC-MS-based metabolite profiling. My results show a clear phenotypic effect of the candidate alpine-adaptive allele. The substrate specificity of the *FAR5* enzyme is shifted from foothill 18C to alpine 16C, resulting in an increased production of a shorter primary fatty alcohol (C16:0-OH) in plants carrying the alpine allele of *FAR5*. This finding experimentally supports *FAR5* as an important player in producing the alpine-adaptive phenotype and brings question about what the function of such metabolomic change is. Thus, I performed a genotype-environment association to unravel which environmental factors might be responsible for the *FAR5* natural variation. I identified an association of the alpine allele of *FAR5* with higher spring precipitation, lower solar radiation in the vegetative season and lower minimum winter temperature. The genome wide association study (GWAS) revealed eight genes significantly associated with similar environmental factors as *FAR5*. Some of these genes are involved in the regulation of seed germination. In connection with the putative effect of the change in substrate specificity on the production of suberin, and *FAR5*'s expression in seed coat, I propose a hypothesis about its adaptive involvement in regulating seed germination. Other factors would also suggest a possible role in pathogen or drought resistance. I suggest that a follow-up targeted study is needed to assess the exact impact of the alpine allele of *FAR5*.

Overall, this study contributes to the understanding of repeated adaptation to the alpine environment through the characterization and functional validation of a candidate alpine-adaptive allele. Unravelling the mechanisms behind such adaptation to dramatic environmental shifts, such as those between lowland and alpine populations, advances our understanding of adaptation. This understanding could be beneficial, for example, in the context of climate change.