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A Genotyping-by-Sequencing approach brings new insights into the population structure and local adaptation of Western Mediterranean Oaks

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The Mediterranean region has been described as a ‘climate change hotspot’, with increased temperatures and decreased precipitation expected to affect the region in the coming decades. Given the pace and intensity at which these changes are expected to happen, it becomes important to understand species capacity to respond to climate change. Species response to environmental change can happen through phenotypic plasticity, range shift, or genetic adaptation to their new conditions. In order to understand a species adaptive capacity to respond to climate change, it is important to disentangle how much of its genetic diversity is the result of population structure, and how much results from the action of natural selection. This work involved samples of Cork Oak (*Quercus suber*) and Holm Oak (*Quercus ilex* and *Quercus rotundifolia*), collected throughout the species range, with special attention given to the Western Mediterranean basin. Genotyping by Sequencing was employed to generate several genome wide Single Nucleotide Polymorphism datasets, which were used (i) to investigate population structure using several complementary approaches, and (ii) to detect evidence of local adaptation through a Landscape Genomics approach involving the detection of genetic-environmental associations with several bioclimatic variables. This work builds on previous analyses of Cork Oak SNP data, and is, to our knowledge, the first attempt to use genome-wide nuclear genetic markers to uncover the

existence of population structure and signatures of local adaptation in Holm oak. Our results reveal contrasting patterns of population structure and differentiation. Holm Oak shows a marked pattern of population structure and considerable differentiation, especially between *Q. rotundifolia* and *Q. ilex* samples, which brings support to the status of *Q. ilex* and *Q. rotundifolia* as two genetically distinct species. Cork Oak, on the other hand, shows much less pronounced population structuring, as reported in previous works based on nuclear genetic markers. Furthermore, a considerable degree of differentiation is observed between Iberian and Moroccan populations of *Q. rotundifolia*, which is not observed for *Q. suber*. Additionally, we uncover the relatively unstructured nature of the Iberian *Q. rotundifolia* and *Q. suber* populations. We also identified a considerable number of putative SNPs under selection in both species, showing association with multiple bioclimatic variables related to temperature and precipitation. Annotation of the genomic regions harboring these putative SNPs revealed several genes potentially associated with heat and water stress. In general, these results build on previous knowledge regarding the population structure of Cork Oak and bring new insights into the population structure of Holm Oak, contributing towards the clarification of its taxonomy, which up until this point has suffered from a lack of consistency. Regarding the detection of local adaptation, our results serve as a first step in understanding the capacity of Cork Oak and Holm Oak to respond to future climate change, opening the door to more complex analyses, such as genomic prediction of maladaptation, which may help identify areas of the species' distribution especially sensitive to climate change, and inform future management efforts towards the conservation of these species.

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