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PROSPECTS FOR DNA BARCODING TO PROVIDE ACCURATE SPECIES
IDENTIFICATION OF *DALBERGIA* (FABACEAE) IN MADAGASCAR

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Prospects for DNA barcoding to provide accurate species identification of *Dalbergia* (Fabaceae) in Madagascar

Alex Widmer & Simon Crameri, ETH Zurich, Zurich, Switzerland

Porter P. Lowry II, Missouri Botanical Garden, St. Louis, MO 63110, USA

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The accurate identification of Malagasy *Dalbergia* species is critical for assessing remaining natural stands, estimating abundance, evaluating conservation status, and informing efforts to control illegal exploitation while promoting sustainable use. Early attempts to use DNA barcoding based on Sanger sequencing of a standard set of chloroplast DNA markers for species identification (Hassold *et al.* 2016) showed the presence of two genetically divergent clades of *Dalbergia* in Madagascar that resulted from at least two colonization events, and which can be readily distinguished by standard DNA barcoding. Within each clade, however, species cannot reliably be distinguished from one another using these techniques. Subsequent work has sought to overcome these limitations by developing a sequence capture approach based on 'anchored phylogenomics' (Lemmon *et al.* 2012) that allows the rapid isolation and analysis of thousands of nuclear DNA fragments using a specially developed high-throughput sequencing technology and bioinformatics pipeline. Our current dataset encompasses 485 samples representing 35 of the 48 described *Dalbergia* species in Madagascar. Key insights from these analyses are that: 1) many of the described taxa form morphologically and geographically coherent and well supported entities, as expected for biological species; 2) at least twelve described taxa (*D. chapelierii*, *D. emirnensis*, *D. hildebrandtii*, *D. lemurica*, *D. maritima*, *D. madagascariensis*, *D. mollis*, *D. monticola*, *D. neoperrieri*, *D. occulta*, *D. pervillei*, *D. purpurascens*), most with apparently large geographic distribution ranges, in fact form more than one entity and likely include hitherto unrecognized new species; and 3) morphological characters or character combinations used for species identification need to be re-interpreted in the light of these new results to inform the re-delimitation of species. Integrating phylogenomic results with morphological analyses of vegetative and reproductive traits is now feasible because herbarium vouchers have been collected for most samples analyzed as part of an effort to establish a comprehensive reference collection for Malagasy *Dalbergia* (ITTO-CITES project TMT-SPD 022/15). Forensic timber identification is also within reach using the 218 fully vouchered heartwood samples collected to date.

Our results offer, for the first time ever, a real prospect for developing a reliable DNA barcoding tool for Malagasy *Dalbergia* species. Such a tool could be used to provide accurate identification of trees regardless of whether they possess the reproductive structures needed for identification. To achieve this goal, the current work would have to be expanded to include multiple samples of all species from throughout their geographic ranges. Beyond the identification of individual trees, the results would also provide valuable input to clarifying species limits among Malagasy *Dalbergia*, reassessing their population and conservation status, developing practical field identification tools, and providing a solid scientific framework for analyses of wood samples (Dormontt *et al.* 2015) with other technologies such as wood morphology (Ravaomanalina *et al.* 2017), near infrared spectroscopy (NIRS), and DART TOF-MS (Espinoza *et al.* 2015). Moreover, our sequence capture approach is applicable across a broad phylogenetic scale and could therefore be readily extended to encompass all *Dalbergia* species worldwide.

References

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