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Research Centre for Ecosystem Resilience (ReCER)

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Foreword

The Research Centre for Ecosystem Resilience (ReCER) is part of the Australian Institute for Botanical Science (AIBS) and it is based at the Royal Botanic Garden Sydney. It was founded in 2020 with a brief to develop innovative science and technology to investigate the factors impacting the distribution and assembly of plant species, and consequently inform the conservation and restoration of resilient ecosystems.

The ReCER team undertakes research and collaborates across multiple areas including genomics, landscape and conservation genetics, ecological restoration, climate modelling, flora biogeography, evolution and microbiome diversity.

The knowledge obtained from our research is disseminated in a number of different ways including academic papers, reports, book chapters and blog posts.

Here we bring together abstracts of publications from the 2022 calendar year, including peer-reviewed publications led or co-authored by the ReCER team. ReCER staff, students or research associate authors are indicated in bold.

Many of our publications are open access and can be downloaded from the links provided below. Blog posts can be read in full at recer.org.au. Other publications and unpublished reports can be requested from the authors.

Peer-reviewed publications

Plant collections for conservation and restoration: Can they be adapted and adaptable?

Jason Bragg, Marlien van der Merwe, Jia-Yee Samantha Yap, Justin Borevitz and Maurizio Rossetto

Molecular Ecology Resources. 22 (6) 2171– 2182

DOI: [10.1111/1755-0998.13605](https://doi.org/10.1111/1755-0998.13605).

Plant collections are important for the conservation of threatened species, and can provide material for ecological restoration. Typically we want collections to have high genetic diversity so populations founded from it are adaptable to future challenges. Sometimes, we have additional objectives for collections, such as enrichment for desirable traits controlled by adaptive alleles. We used landscape genomic datasets for two plants, *Westringia fruticosa* and *Wilkiea huegeliana*, to design collections that are genetically diverse, and that are adapted to warming climates. We characterized temperature adaptation by: (i) using the mean annual temperature of the sites of origin of the plants, and (ii) using the representation of alleles that are associated with warm temperatures. In *Westringia fruticosa*, there was a negative correlation, or tradeoff, between designing a collection that was both genetically diverse and adapted to warm temperatures. This tradeoff was weaker in *Wilkiea huegeliana*. We hypothesized this was because neutral genetic variation was strongly correlated with temperature in *Westringia fruticosa*, and not in *Wilkiea huegeliana*. Accordingly, when we shuffled the temperature data, breaking up the covariance between *Westringia fruticosa* genetic variation and temperature, there was a relaxation of the observed tradeoff. In sum, we explore tradeoffs between promoting genetic diversity and selecting for a specific trait in plant collections, and show that the strength of this tradeoff varies between two species. This represents a useful step towards understanding when selection will have a large cost in genetic diversity, and when it will be possible to design a collection that is both adapted and adaptable.



Phylogenetic Reconstruction of the Rainforest Lineage *Fontainea* Heckel (Euphorbiaceae) Based on Chloroplast DNA Sequences and Reduced-Representation SNP Markers.

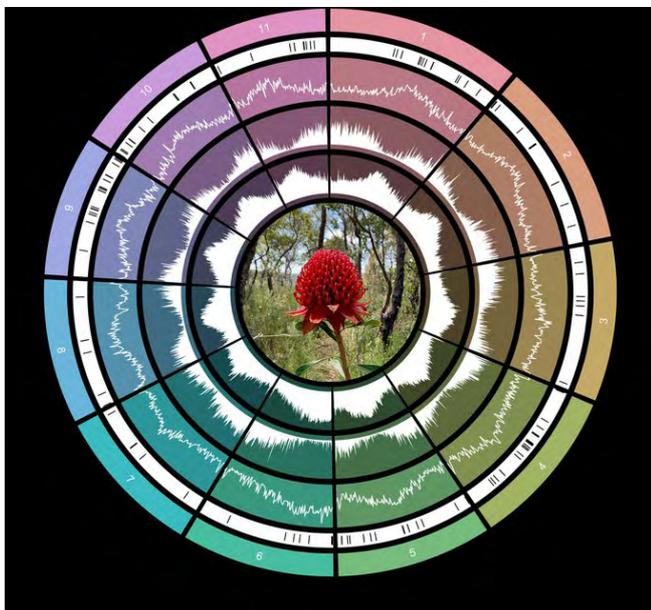
Aaron Brunton, Robert Lamont, Gabriel Conroy, **Jia-Yee Samantha Yap, Maurizio Rossetto**, Alyce Taylor-Brown, Laurent Maggia, Paul Reddell and Steven Ogbourne.

Diversity 14, 725

DOI: [10.3390/d14090725](https://doi.org/10.3390/d14090725).

Fontainea is a plant genus with nine recognised species that occur across the tropical and subtropical rainforests of Australia, Papua New Guinea, New Caledonia, and Vanuatu. One of these species is cultivated commercially as the source of a cancer therapeutic, and several other species are under threat of extinction. Despite this, the phylogenetic relationships of the genus have not been explored. Our study assessed the phylogeny of seven *Fontainea* taxa from the Australian and Pacific Island complex using chloroplast DNA sequence data and reduced-representation genome sequencing. Maximum-likelihood and consensus network trees were used to infer the topology of phylogenetic relationships between species, which highlighted three distinct lineages and a number of sister species. Our results indicated that the geographically disjunct species *Fontainea venosa* and *F. pancheri* formed a sister group at the earliest position of divergence for the genus. The data also revealed

that the vulnerable *Fontainea australis* and the critically endangered *F. oraria* form a sister subclade with evidence of some shared plastid genotypes. Generally, our phylogenetic reconstruction supports the modern taxonomical nomenclature. However, we suggest further accessions across several species may support improved genetic distinctions between the sister groups of *Fontainea* within the genus.



Chromosome-level de novo genome assembly of *Telopea speciosissima* (New South Wales waratah) using long-reads, linked-reads and Hi-C.

Stephanie Chen, Maurizio Rossetto, Marlien van der Merwe, Patricia Lu-Irving, Jia-Yee Samantha Yap, Hervé Sauquet, Greg Bourke, Jason Bragg and Richard Edwards.

Molecular Ecology Resources. 22 (5) 1836-1854

DOI: [10.1111/1755-0998.13574](https://doi.org/10.1111/1755-0998.13574).

Background: *Telopea speciosissima*, the New South Wales waratah, is Australian endemic woody shrub in the family Proteaceae. Waratahs have great potential as a model clade to better understand processes of speciation, introgression and adaptation, and are significant from a horticultural perspective.

Findings: Here, we report the first chromosome-level reference genome for *T. speciosissima*. Combining Oxford Nanopore long-reads, 10x Genomics Chromium linked-reads and Hi-C data, the assembly spans 823 Mb (scaffold N50 of 69.0 Mb) with 91.2 % of Embryophyta BUSCOs complete. We introduce a new method in Diploidocus (<https://github.com/slimsuite/diploidocus>) for classifying, curating and QC-filtering assembly scaffolds. We also present a new tool, DepthSizer (<https://github.com/slimsuite/depthsizer>), for genome size estimation from the read depth of single copy orthologues and find that the assembly is 93.9 % of the estimated genome size. The largest 11 scaffolds contained 94.1 % of the assembly, conforming to the expected number of chromosomes ($2n = 22$). Genome annotation predicted 40,158 protein-coding genes, 351 rRNAs and 728 tRNAs. Our results indicate that the waratah genome is highly repetitive, with a repeat content of 62.3 %.

Conclusions: The *T. speciosissima* genome (Tspe_v1) will accelerate waratah evolutionary genomics and facilitate marker assisted approaches for breeding. Broadly, it represents an important new genomic resource of Proteaceae to support the conservation of flora in Australia and further afield.

Applying biocultural research protocols in ecology: Insider and outsider experiences from Australia.

Patrick Cooke, **Monica Fahey**, Emilie J. Ens, Margaret Raven, Philip Allan Clarke, **Maurizio Rossetto** and Gerry Turpin

Ecological Management & Restoration. 23. 64-74

DOI: [10.1111/emr.12545](https://doi.org/10.1111/emr.12545).

Collaborations between Indigenous and non-Indigenous scientific researchers are increasingly mandated by global to local conservation policy and research ethics guidelines. Breakdowns occur due to misunderstandings around expected protocols of engagement and cooperation, which are compounded by lack of broader awareness of differences in cultural values, priorities and knowledge systems. Using first-hand experiences, we outline eight key protocols and guidelines that researchers should consider when undertaking research with Indigenous peoples, or on Indigenous Country, through exploration of biocultural protocols and guidelines within Australian and Indigenous customary laws. We use the onion as a metaphor to highlight the layers of protocols and guidelines that researchers can peel back to guide their research from international to local scales, with ethics around the research question at the core. This paper draws on the perspectives and experiences of an Indigenous researcher (as 'insider'/'outsider') and non-Indigenous researcher ('outsider'), working on a cross-cultural and multidisciplinary investigation of past Aboriginal dispersal of rainforest trees on the Australian east coast. This paper is part of the special issue 'Indigenous and cross-cultural ecology - perspectives from Australia' published in Ecological Management & Restoration.

Genomic Screening to Identify Food Trees Potentially Dispersed by Precolonial Indigenous Peoples.

Monica Fahey, Maurizio Rossetto, Emilie J. Ens and Andrew Ford

Genes. 13. 476.

DOI: [10.3390/genes13030476](https://doi.org/10.3390/genes13030476).

Over millennia, Indigenous peoples have dispersed the propagules of non-crop plants through trade, seasonal migration or attending ceremonies; and potentially increased the geographic range or abundance of many food species around the world. Genomic data can be used to reconstruct these histories. However, it can be difficult to disentangle anthropogenic from non-anthropogenic dispersal in long-lived non-crop species. We developed a genomic workflow that can be used to screen out species that show patterns consistent with faunal dispersal or long-term isolation, and identify species that carry dispersal signals of putative human influence. We used genotyping-by-sequencing (DArTseq) and whole-plastid sequencing (SKIMseq) to identify nuclear and chloroplast Single Nucleotide Polymorphisms in east Australian rainforest trees (4 families, 7 genera, 15 species) with large (>30 mm) or small (<30 mm) edible fruit, either with or without a known history of use by Indigenous peoples. We employed standard population genetic analyses to test for four signals of dispersal using a limited and opportunistically acquired sample scheme. We expected different patterns for species that fall into one of three broadly described dispersal histories: (1) ongoing faunal dispersal, (2) post-megafauna isolation and (3) post-megafauna isolation followed by dispersal of putative human influence. We identified five large-fruited species that displayed strong population structure combined with signals of dispersal. We propose coalescent methods to investigate whether these genomic signals can be attributed to post-megafauna isolation and dispersal by Indigenous peoples.



▼
A box on the river: The phylogenetics and phylogeography of *Eucalyptus baueriana* (*Eucalyptus* sect. *Adnataria* ser. *Heterophloiae*).

Patrick Fahey, Frank Udovicic, David J Cantrill and Michael Bayly

PLOS ONE. 17. e0276117.

DOI: [10.1371/journal.pone.0276117](https://doi.org/10.1371/journal.pone.0276117)

We present a phylogeographic study of the tree species *Eucalyptus baueriana* Schauer, which occurs in disjunct areas on the near coastal plains and ranges of the south-east Australian mainland. DArTseq data are used to build a phylogeny including *E. baueriana* and closely related taxa to test its monophyly, test the genetic distinctness of the three subspecies of *E. baueriana*, and investigate relationships between its disjunct populations. Additionally, we use population structure analysis to investigate the genetic distinctness of populations, and MaxEnt to investigate the environmental factors potentially influencing the species' distribution. We show *E. baueriana* is monophyletic and most closely related to three other Blue Box eucalypt species: *E. conica* H.Deane & Maiden, *E. dalveenica* T.L.Collins, R.L.Andrew & J.J.Brühl and *E. magnificata* L.A.S.Johnson & K.D.Hill, with some evidence for genetic introgression between these taxa. Within *E. baueriana*, the deepest genetic breaks do not correspond with the subspecies classification as the two geographically restricted subspecies, together with samples of the more widespread *E. baueriana* subsp. *baueriana* from west of the Gippsland lowlands, form a south-western clade with that is sister to other populations of subsp. *baueriana*. The oldest genetic break in the species occurs in far eastern Gippsland (Victoria), corresponding to one of the shortest geographic disjunctions in the species' distribution. Genetic breaks in other species have been observed in this region which is broadly referred to as the southern transition zone. Both total annual rainfall and the seasonality of this rainfall are hypothesised to affect the species' distribution; gaps in its distribution are in areas of higher rainfall that support closed forest and in regions with more winter dominated rainfall.

A phylogenetic investigation of the taxonomically problematic *Eucalyptus odorata* complex (*E.* section *Adnataria* series *Subbuxuales*): evidence for extensive interspecific gene flow and reticulate evolution.

Patrick Fahey, Frank Udovicic, David J Cantrill, Dean Nicolle, Todd Mclay and Michael Bayly

Australian Systematic Botany. 35. 403.

DOI: [10.1071/SB21029](https://doi.org/10.1071/SB21029)

To investigate the relationships among species in the taxonomically problematic *Eucalyptus odorata* species complex, we generated molecular data using double-digest restriction site-associated DNA

sequencing (ddRADseq) and Diversity Arrays Technology sequencing (DArTseq). These data were analysed utilising principal-component analysis (PCA), phylogenetic networks, phylogeny reconstruction and hybridisation tests. Twelve species that are variously recognised in the complex were sampled from across their ranges, along with co-occurring members of *E.* section *Adnataria*, to allow for patterns of hybridisation and gene flow to be identified. Despite the large genetic datasets generated, many relationships within the *E. odorata* complex were poorly resolved, and few species were monophyletic, likely owing to both biological factors including recent speciation and extensive hybridisation and introgression, and potential over-splitting of taxa. We show that multiple taxa with limited distributions are the result of reticulate evolutionary events and that typical *Eucalyptus viridis* R.T.Baker and the possibly con-specific *E. aenea* K.D.Hill are sister to the rest of the complex. The remaining species appeared to represent a discontinuous crescent-shaped cline running from the Flinders Ranges to the south-western slopes region of New South Wales, with limited support for an east-west split in this cline across the Murray River Basin. *Eucalyptus viridis* var. *latiuscula* Blakely, which is not closely related to the typical variety of this species in our data, may represent a northern extension to this cline.

Global genetic diversity status and trends: towards a suite of Essential Biodiversity Variables (EBVs) for genetic composition

Sean Hoban, Frederick Ivan Archer, Laura Bertola, **Jason Bragg** et al.

Biological Reviews. 97 (4) 1511-1538.

DOI: [DOI:10.1111/brv.12852](https://doi.org/10.1111/brv.12852)

Biodiversity underlies ecosystem resilience, ecosystem function, sustainable economies, and human well-being. Understanding how biodiversity sustains ecosystems under anthropogenic stressors and global environmental change will require new ways of deriving and applying biodiversity data. A major challenge is that biodiversity data and knowledge are scattered, biased, collected with numerous methods, and stored in inconsistent ways. The Group on Earth Observations Biodiversity Observation Network (GEO BON) has developed the Essential Biodiversity Variables (EBVs) as fundamental metrics to help aggregate, harmonize, and interpret biodiversity observation data from diverse sources. Mapping and analyzing EBVs can help to evaluate how aspects of biodiversity are distributed geographically and how they change over time. EBVs are also intended to serve as inputs and validation to forecast the status and trends of biodiversity, and to support policy and decision making. Here, we assess the feasibility of implementing Genetic Composition EBVs (Genetic EBVs), which are metrics of within-species genetic variation. We review and bring together numerous areas of the field of genetics and evaluate how each contributes to global and regional genetic biodiversity monitoring with respect to theory, sampling logistics, metadata, archiving, data aggregation, modeling, and technological advances. We propose four Genetic EBVs: (i) Genetic Diversity; (ii) Genetic Differentiation; (iii) Inbreeding; and (iv) Effective Population Size (Ne). We rank Genetic EBVs according to their relevance, sensitivity to change, generalizability, scalability, feasibility and data availability. We outline the workflow for generating genetic data underlying the Genetic EBVs, and review advances and needs in archiving genetic composition data and metadata. We discuss how Genetic EBVs can be operationalized by visualizing EBVs in space and time across species and by forecasting Genetic EBVs beyond current observations using various modeling approaches. Our review then explores challenges of aggregation, standardization, and costs of operationalizing the Genetic EBVs, as well as future directions and opportunities to maximize their uptake globally in research and policy.

The collection, annotation, and availability of genetic data has made major advances in the past decade, each of which contributes to the practical and standardized framework for large-scale genetic observation reporting. Rapid advances in DNA sequencing technology present new opportunities, but also challenges for operationalizing Genetic EBVs for biodiversity monitoring regionally and globally. With these advances, genetic composition monitoring is starting to be integrated into global conservation policy, which can help support the foundation of all biodiversity and species' long-term persistence in the face of environmental change. We conclude with a summary of concrete steps for researchers and policy makers for advancing operationalization of Genetic EBVs. The technical and analytical foundations of Genetic EBVs are well developed, and conservation practitioners should anticipate their increasing application as efforts emerge to scale up genetic biodiversity monitoring regionally and globally.

Threatened Species Initiative: Empowering conservation action using genomic resources.

Carolyn J. Hogg, Kym Ottewell, Peter Latch, **Maurizio Rossetto**, James Biggs, Andrew Gilbert, Sarah Richmond and Katherine Belov
Proceedings of the National Academy of Sciences. 119 (4) e2115643118.
DOI: [10.1073/pnas.2115643118](https://doi.org/10.1073/pnas.2115643118).

Globally, 15,521 animal species are listed as threatened by the International Union for the Conservation of Nature, and of these less than 3% have genomic resources that can inform conservation management. To combat this, global genome initiatives are developing genomic resources, yet production of a reference genome alone does not conserve a species. The reference genome allows us to develop a suite of tools to understand both genome-wide and functional diversity within and between species. Conservation practitioners can use these tools to inform their decision-making. But, at present there is an implementation gap between the release of genome information and the use of genomic data in applied conservation by conservation practitioners. In May 2020, we launched the Threatened Species Initiative and brought a consortium of genome biologists, population biologists, bioinformaticians, population geneticists, and ecologists together with conservation agencies across Australia, including government, zoos, and nongovernment organizations. Our objective is to create a foundation of genomic data to advance our understanding of key Australian threatened species, and ultimately empower conservation practitioners to access and apply genomic data to their decision-making processes through a web-based portal. Currently, we are developing genomic resources for 61 threatened species from a range of taxa, across Australia, with more than 130 collaborators from government, academia, and conservation organizations. Developed in direct consultation with government threatened-species managers and other conservation practitioners, herein we present our framework for meeting their needs and our systematic approach to integrating genomics into threatened species recovery.



Gondwanan survivor lineages and the high-risk biogeography of Anthropocene Southeast Asia.

Robert Kooyman, Sarah Ivory, Adam Benfield and Peter Wilf
Journal of Systematics and Evolution. 60 (4) 715-727
DOI: [10.1111/jse.12853](https://doi.org/10.1111/jse.12853).

The Southeast Asian rainforest region is extremely complex and biodiverse. Fossils have shown that paleo-Antarctic rainforest lineages (PARLs) now extant in Asia tracked the ever-wet conditions needed to survive and diversify through deep time. However, the threat of future climate change to the remaining rainforest and PARLs in Southeast Asia has yet to be evaluated to set conservation priorities. We first quantified the woody-genus floristic relationships of Southeast Asian Island Groups by vetting and analyzing recent compilations of bioregional species data. We then evaluated the contributions to community assembly of woody fossil lineages and Island Group relationships to environmental gradients. To better understand climatic constraints of fossil lineage distributions and forecast distributions under projected future climate, we used exemplar living woody PARLs, including two angiosperms and two gymnosperms. Generalized linear models were used to project potential distributions under future climate pathways that assume no reduction in carbon dioxide emissions. The floristic analyses highlighted strong similarity among Island Groups in the ever-wet forest areas of Malesia, where PARLs are often concentrated. Ordination outliers represented more seasonal locations. Species distribution models showed that potential future distributions of ancient lineages are constrained by increasing rainfall seasonality and higher seasonal temperatures, with significant differences among exemplar genera. Notably, potential distributions often mapped onto de facto inaccessible areas, where forest clearing and the ubiquitous marine dispersal barriers that characterize the region will drastically inhibit potential relocation. These realities gravely threaten paleo-conservation values and contemporary rainforest community assembly processes in Southeast Asia.

New insights from population genomics into the invasive *Lantana camara* L species complex.

Pat Lu-Irving, F Encinas-Viso, J Callander, MD Day, JJ Le Roux
Peer-reviewed proceedings of the 22nd Australasian Weeds Conference. Council of Australasian Weed Societies pp. 45–47.

Extensive morphological diversity in the invasive *L. camara* species complex has resisted clear taxonomic characterisation, yet molecular studies to date have revealed minimal genetic structure. Analysis of thousands of genome-wide markers successfully detects differentiation among sub-lineages within the complex, revealing that two of the morphological variants in Australia comprise multiple, distinct lineages. The common pink flowering morphotype appears to be monophyletic, whereas the pink-edged red flowering morphotype does not. Pending further investigation and expanded sampling, these findings hold promise for improving weed management through a deeper understanding of the systematics of the *L. camara* complex (e.g., by enabling selection of biological control agents that are best matched with target host populations).



The culturable seed mycobiome of two *Banksia* species is dominated by latent saprotrophic and multi-trophic fungi.

Allison Mertin, Matthew Laurence, Marlien van der Merwe, Kris French and Edward Liew

Fungal Biology, 126, (11–12) 738–745

DOI: [10.1016/j.funbio.2022.09.002](https://doi.org/10.1016/j.funbio.2022.09.002)

Seed fungal endophytes play an important beneficial role in the formation of the seedling mycobiome and contribute to plant establishment, but can also occur as latent pathogens and saprotrophs. Current knowledge on the function and diversity of seed fungal endophytes has been gained through studies in agricultural systems whilst knowledge from natural systems is relatively less. We used two co-occurring species from the genus *Banksia* from four sites in Australia's Sydney Basin Bioregion to investigate the abundance and diversity of seed fungal endophyte communities present in natural ecosystem hosts. Based on results from culturing and DNA sequence analysis of multiple loci, we found that *Banksia* seeds house a diverse range of fungal endophyte species, that when assigned to functional guilds belonged to multiple trophic modes. Thirty-one of the fungal taxa identified had not been previously reported as endophytes. Amongst the 58 Operational Taxonomic Units identified, Leotiomyces and Sordariomyces were the dominant classes and *Banksiamyces* (Leotiomyces) and *Penicillium* (Sordariomyces) the dominant genera, with many of the species isolated recorded in the literature as having a limited distribution. The two *Banksias* shared few fungal endophyte species, which were not always present across all study sites. We revealed a 'hidden diversity' within seeds of *Banksia* from natural ecosystems and provided insights into the influence host species can have on the seed mycobiome.

Patterns and drivers of plant diversity across Australia.

Karel Mokany, James McCarthy, Daniel Falster, Rachael Gallagher, Thomas D. Harwood, Robert Kooyman and Mark Westoby

Ecography, 11, e06426

DOI: [10.1111/ecog.06426](https://doi.org/10.1111/ecog.06426)

Biodiversity analyses across continental extents are important in providing comprehensive information on patterns and likely drivers of diversity. For vascular plants in Australia, community-level diversity analyses have been restricted by the lack of a consistent plot-based survey dataset across the continent. To overcome these challenges, we collated and harmonised plot-based vegetation survey data from the major data sources across Australia and used

them as the basis for modelling species richness (α -diversity) and community compositional dissimilarity (β -diversity), standardised to 400 m², with the aim of mapping diversity patterns and identifying potential environmental drivers. The harmonised Australian vegetation plot (HAVPlot) dataset includes 219 552 plots, of which we used 115 083 to analyse plant diversity. Models of species richness and compositional dissimilarity both explained approximately one-third of the variation in plant diversity across Australia ($D_2 = 33.0\%$ and 32.7% , respectively). The strongest environmental predictors for both aspects of diversity were a combination of temperature and precipitation, with soil texture and topographic heterogeneity also important. The fine-resolution (≈ 90 m) spatial predictions of species richness and compositional dissimilarity identify areas expected to be of particular importance for plant diversity, including south-western Australia, rainforests of eastern Australia and the Australian Alps. Arid areas of central and western Australia are predicted to support assemblages that are less speciose or unique; however, these areas are most in need of additional survey data to fill the spatial, environmental and taxonomic gaps in the HAVPlot dataset. The harmonised data and model predictions presented here provide new insight into plant diversity patterns across Australia, enabling a wide variety of future research, such as exploring changes in species abundances, linking compositional patterns to functional traits or undertaking conservation assessments for selected components of the flora.

Evolutionary processes in an undescribed eucalypt: implications for the translocation of a critically endangered species.

Susan Rutherford, Trevor Wilson, Samantha Yap, Enhua Lee, Graeme Errington and Maurizio Rossetto

Annals of Botany, Volume 130, Issue 4

DOI: [10.1093/aob/mcac091](https://doi.org/10.1093/aob/mcac091)

Background and Aims: Knowledge of the evolutionary processes responsible for the distribution of threatened and highly localised species is important for their conservation. Population genomics can provide insights into evolutionary processes to inform management practices, including the translocation of threatened plant species. In this study, we focus on a critically endangered eucalypt, *Eucalyptus* sp. Cattai, which is restricted to a 40 km² area of Sydney, Australia and is threatened by increased urbanisation. *Eucalyptus* sp. Cattai is yet to be formally described in part due to its suspected hybrid origin. Here, we examined evolutionary processes and species boundaries in *E. sp. Cattai* to determine whether translocation was warranted. Methods: We used genome-wide scans to investigate the evolutionary relationships of *E. sp. Cattai* with related species, and to assess levels of genetic health and admixture. Morphological trait and genomic data were obtained from seedlings of *E. sp. Cattai* propagated in a common garden to assess their genetic provenance and hybrid status. Key Results: All analyses revealed that *E. sp. Cattai* was strongly supported as a distinct species. Genetic diversity varied across populations, and clonality was unexpectedly high. Interspecific hybridisation was detected, and was more prevalent in seedlings compared to in situ adult plants, indicating that post-zygotic barriers may restrict the establishment of hybrids. Conclusions: Multiple evolutionary processes (e.g., hybridisation and clonality) can operate within the one rare and restricted species. Insights regarding evolutionary processes from our study were used to assist with the translocation of genetically 'pure' and healthy ex situ seedlings to nearby suitable habitat. Our findings demonstrate that it is vital to provide an understanding of evolutionary relationships and processes with an examination of population genomics in the design and implementation of an effective translocation strategy.

Detecting steps in spatial genetic data: Which diversity measures are best?

Alexander Sentinella, Angela Moles, **Jason Bragg**, **Maurizio Rossetto**, and William Bruce Sherwin

PloS one. 17(3): e0265110

DOI: [10.1371/journal.pone.0265110](https://doi.org/10.1371/journal.pone.0265110).

Accurately detecting sudden changes, or steps, in genetic diversity across landscapes is important for locating barriers to gene flow, identifying selectively important loci, and defining management units. However, there are many metrics that researchers could use to detect steps and little information on which might be the most robust. Our study aimed to determine the best measure/s for genetic step detection along linear gradients using biallelic single nucleotide polymorphism (SNP) data. We tested the ability to differentiate between linear and step-like gradients in genetic diversity, using a range of diversity measures derived from the q-profile, including allelic richness, Shannon Information, GST, and Jost-D, as well as Bray-Curtis dissimilarity. To determine the properties of each measure, we repeated simulations of different intensities of step and allele proportion ranges, with varying genome sample size, number of loci, and number of localities. We found that alpha diversity (within-locality) based measures were ineffective at detecting steps. Further, allelic richness-based beta (between-locality) measures (e.g., Jaccard and Sørensen dissimilarity) were not reliable for detecting steps, but instead detected departures from fixation. The beta diversity measures best able to detect steps were: Shannon Information based measures, GST based measures, a Jost-D related measure, and Bray-Curtis dissimilarity. No one measure was best overall, with a trade-off between those measures with high step detection sensitivity (GST and Bray-Curtis) and those that minimised false positives (a variant of Shannon Information). Therefore, when detecting steps, we recommend understanding the differences between measures and using a combination of approaches.

A turn in species conservation for hairpin banksias: demonstration of oversplitting leads to better management of diversity.

Trevor Wilson, **Maurizio Rossetto**, David Bain, **Jia-Yee Samantha Yap**, **Peter D. Wilson**, Margaret Stimpson, Peter Weston and Larry Croft.

American Journal of Botany. 109 (10)

DOI: [10.1002/ajb2.16074](https://doi.org/10.1002/ajb2.16074).

Premise: Understanding evolutionary history and classifying discrete units of organisms remains an overwhelming task, and lags in this discipline concomitantly impede an accurate documentation of biodiversity and conservation management. Rapid advances and improved accessibility of sensitive high throughput sequencing tools are fortunately quickening the resolution of morphological complexes that generally are underestimating species diversity. This seems to be the case of the persisting taxonomic quandary of the Hairpin Banksias (*B. spinulosa* sens. lat.), a group of eastern Australian flowering shrubs demonstrating a continuum of morphological diversity from which the critically endangered *B. vincentia* has been described. Methods: To assist conservation while testing the current taxonomy of this group, high-throughput sequencing was used to infer a population-scale evolutionary scenario for a sample-set comprehensive in its representation of morphological diversity and a two-and-a-half thousand kilometer distribution. Key results: *Banksia spinulosa* sens. lat. represents two clades, each with an internal genetic structure shaped through historical separation by biogeographic barriers. This structure conflicts with the existing taxonomy for the group. Corroboration between phylogeny and population statistics aligns with

the hypothesis that *B. collina*, *B. neoanglica*, and *B. vincentia* should not be classified as species. Conclusions: The pattern here supports how morphological diversity can be indicative of a locally expressed suite of traits rather than relationship. Over-splitting in the Hairpin Banksias is atypical since genomic analyses often reveal that species diversity is underestimated. However, we show that erring on overestimation can yield negative consequences, such as the disproportionate prioritisation of a geographically anomalous population.

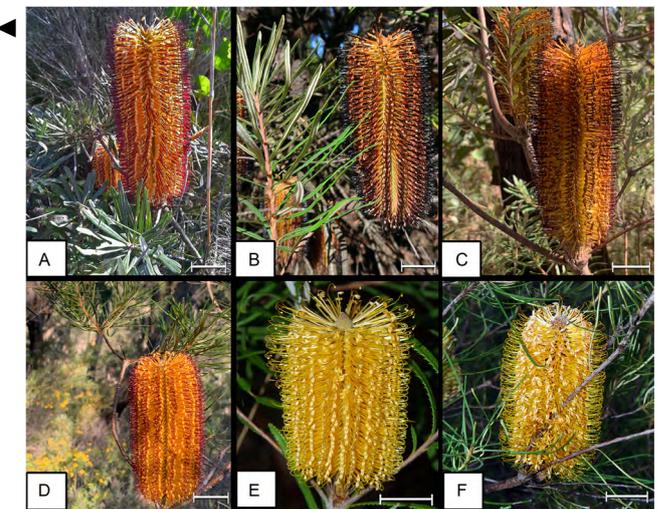
Tracking habitat or testing its suitability? Similar distributional patterns can hide very different histories of persistence vs non-equilibrium dynamics.

Jia-Yee Samantha Yap, **Maurizio Rossetto**, Sourav Das, **Peter D. Wilson**, Linda Beaumont and Robert Henry

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The expansions and contractions of a species' range in response to temporal changes in selective filters leave genetic signatures that can inform a more accurate reconstruction of their evolutionary history across the landscape. After a long period of continental decline, Australian rainforests settled into localised patterns of contraction / expansion during the climatic fluctuations of the Quaternary. The environmental impacts of recurring glacial and interglacial periods also intensified the arrival of new lineages from the Sunda shelf, and it can be expected that immigrant vs locally persistent taxa responded to environmental challenges in quantifiably different manner. To investigate how such differences impact on species' distribution, we contrast landscape genomic patterns and changes in habitat availability between a species with a long continental history on (*Doryphora sassafras*) and a Sunda-derived species (*Toona ciliata*), across a distributional overlap. Extensive landscape-level homogeneity across chloroplast and nuclear genomes for the Sunda-derived *T. ciliata*, characterize the genetic signature of a very recent invasion and a rapid southern 'exploratory' expansion that had not been previously recorded in the Australian flora (i.e. of Gondwanan origin or Sahul-derived). In contrast, *D. sassafras* is consistent with other Sahul-derived species characterized by strong geographical divergence and regional differentiation. Interestingly, our findings suggest that admixture between genetically divergent populations during expansion events might be a contributing factor to the successful colonization of novel habitats. Overall, this study identifies some of the mechanisms regulating the rearrangements in species distributions and assemblage composition that follow major environmental shifts, and reminds us how a species' current range might not necessarily define species' habitat preference, with the consequence that estimates of past / future range might not always be reliable.



Unpublished reports prepared for the NSW Saving our Species program

Conservation genomics of *Acacia terminalis* subsp. Eastern Sydney in support of management.

Jia-Yee Samantha Yap, Marlien van der Merwe and Maurizio Rossetto.

Taxonomic status and genetic health of an undescribed but listed as endangered subspecies of *Acacia terminalis*, Eastern Sydney, was assessed using high quality genome scans. We provide empirical evidence confirming its subspecies status. We show that *A. terminalis* subsp. Eastern Sydney is morphologically variable but hybridisation was not detected among individuals sampled. Multiple *Acacia terminalis* populations neighbouring the currently described distribution of this rare subspecies were genetically similar to it, indicating the geographic extent of subsp. Eastern Sydney is larger than described. An in-depth study on individuals within the Sydney region will be needed to clearly define the true extent of subsp. Eastern Sydney and guide adequate management strategies accordingly. Infra-subspecific genetic diversity is present within the taxa that includes subsp. Eastern Sydney and neighbouring populations. Lower levels of diversity in the populations of subsp. Eastern Sydney indicate fragmentation impacted on their highly restricted distribution. Assuring maximum levels of genetic diversity in an ex-situ collection will increase fitness, reducing the risk of inbreeding as well as increasing adaptive potential to environmental change and other pressures. We calculated the necessary combinations of propagules for multiple scenarios that will ensure the establishment of suitably evolutionary resilient ex-situ collections and plantings.

Stage 2 Conservation genomics of *Astrotricha crassifolia* in support of management and translocation activities.

Eilish McMaster, Richard Dimon, Jia-Yee Samantha Yap and Maurizio Rossetto.

A conservation genomics study on *Astrotricha crassifolia* conducted in 2020 (Stage 1) found the taxa to be more restricted than previously believed, with only individuals at Brisbane Water National Park representing the bona fide species. Specimens identified elsewhere are characterised by extensive hybridisation. The Brisbane Water population although genetically variable, has lower diversity relative to comparative populations from other *Astrotricha* species, reflecting the species' vulnerable status. Furthermore, hybridisation has also occurred at the Brisbane Water site, suggesting that *A. crassifolia* is at high risk of total genetic swamping. Since the 2020 study, surveys were carried out to locate further individuals and this follow-up stage 2 study was proposed to ensure all pure individuals are known and actively managed. All additional individuals of *A. crassifolia* sampled from Brisbane Water NP in this study were pure. Optimised scenarios representing selection of individuals to be included in representative ex-situ collections that capture diversity within the remaining population are provided.

Conservation genomics of *Bossiaea* species in South-eastern Australia in support of management.

Eilish McMaster, Jia-Yee Samantha Yap and Maurizio Rossetto.

This genomics report was ordered to investigate the phylogeny and genetic health of the rare *Bossiaea* species *B. fragrans*, *B. milesiae*, *B.*

bombayensis, and *B. grayi* found in south-eastern New South Wales and the Australian Capital Territory. These species were found to be taxonomically distinct with no evidence of recent hybridisation, however low levels of genetic segregation indicate cross breeding may be possible. There are high levels of relatedness and clonality within all four species, with *B. milesiae* of particular concern due to extensive clonality. As a result, genetic diversity is low in all populations, making these species highly susceptible to disturbance and collapse. Protection from further population fragmentation is essential to preventing further decline. Strategic classification and intermixing of closely related *B. fragrans* and *B. milesiae* should be considered to maximise genetic diversity and chance of survival.

Conservation genomics of *Lenwebbia* sp. Main Range in support of management.

Stephanie Chen, Jia-Yee Samantha Yap, Jason Bragg and Maurizio Rossetto.

The genetic diversity and structure of *Lenwebbia* sp. Main Range (Myrtaceae), which is critically endangered due to myrtle rust, were measured using high quality genome scans (DARtseq). We find that *L. sp. Main Range* is not genetically distinct from all other similarly distributed *Lenwebbias*, namely *L. sp. Blackall Range* and *L. prominens*, and that genetically intermediate individuals are present in the wild. Therefore, it is recommended that the species complex should be managed as a whole. Genetic diversity within the complex is distributed along a longitudinal gradient, and while current ex situ collections at the Australian Botanic Garden Mount Annan adequately captures the genetic diversity of *L. sp. Main Range*, they should expand to include other *Lenwebbias*. Kinship analyses revealed some clonality among individuals in the collection, suggesting that identification of genets is sometimes problematic, and that a follow-up genetic study would be helpful to ensure maximal genetic diversity and minimal kinship is maintained in the collection when other *Lenwebbias* are incorporated. To inform the selection of individuals for specific management actions, future studies should investigate the extent of genetic diversity in the species complex by comprehensively sampling across the distribution of *L. sp. Blackall Range* and *L. prominens*; *L. lasioclada* should also be included to ascertain if it is part of the species complex since *L. sp. Main Range* was previously described as part of *L. lasioclada*. The maximisation of genetic diversity will ensure the collection is viable in the long-term by reducing the risk of inbreeding and maintaining high levels of adaptive potential to environmental change and other pressures. The genetic data and recommendations from this study will facilitate the onground management of *Lenwebbia* to contribute to conservation management.



Conservation genomics of *Pimelea venosa* and *Pimelea cremnophila* in support of management.

Eilish McMaster, Jia-Yee Samantha Yap and Maurizio Rossetto.

This study was conducted to determine the population structure and health of the endangered species *Pimelea venosa* and related critically endangered species *Pimelea cremnophila*. Both species displayed low levels of heterozygosity and high inbreeding coefficients, that was similarly observed in other species, suggesting the Pimeleas may be prone to inbreeding when populations experience fragmentation and isolation. For the extremely rare and localised, *P. cremnophila*, the presence of genetic diversity indicates surveys are needed to further locate populations/diversity so that the species can be better managed as a whole. For *P. venosa*, an extremely limited gene flow between populations comparable to between species and a recent sprouting of *P. venosa* seedlings from a soil seedbank, provided further insights into how genetic diversity is currently being maintained and how to manage it to ensure long-term survival. For example, we recommend the inclusion of individuals from one population in the other population to increase genetic diversity (and thus general fitness of the populations). To add to this, we calculated the necessary combinations of propagules for scenarios that will ensure the establishment of suitably resilient ex-situ collections for future plantings.

Conservation genomics of *Pomaderris delicata* in support of management.

Stephanie Chen, Jia-Yee Samantha Yap and Maurizio Rossetto

The genetic diversity and structure of *Pomaderris delicata* (Rhamnaceae), which is critically endangered and known from only two sites, were measured using high quality genome scans (DArTseq). We find that *P. delicata* clusters away from *P. andromedifolia* which was used as the outgroup. However, there is still taxonomic uncertainty as only one *P. delicata* genet was sampled, so we cannot conclude if *P. delicata* is genetically distinct. Due to the very low level of heterozygosity in *P. delicata*, we were unable to confirm the ploidy status of the sampled individuals, although the outgroup was confirmed to be a polyploid. Genetic diversity within the species is very limited and there is low heterozygosity, and this is likely a result of inbreeding across all populations. The genetic data and recommendations from this study will facilitate the on-ground management of *Pomaderris delicata* and contribute to conservation management.

Conservation genomics of *Pultenaea maritima* in support of management.

Eilish McMaster, Maurizio Rossetto and Jia-Yee Samantha Yap.

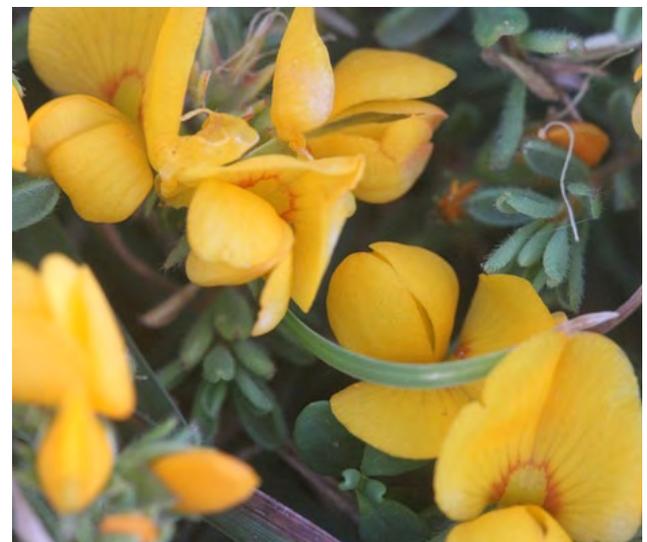
This study was conducted to determine whether the vulnerable species *Pultenaea maritima* is distinct from the common species *Pultenaea villosa*, to assess the population health of *P. maritima*, and to test if there is a genetic basis for morphology variation observed. Samples of *P. maritima* were collected from subpopulations along the NSW coast from Evans Head to Pinny beach. The genetic analyses found low levels of genetic diversity in all subpopulations of *P. maritima*, and all subpopulations also present detectable levels of inbreeding likely due to selfing. Inbreeding and limited gene flow between geographically separated sites has resulted in highly structured

subpopulations and genetic isolation between northern and southern subpopulations, which may account for phenotypic differences observed. The southern group is not phylogenetically distinct or genetically segregating from *P. villosa*, indicating they are the same species. Based on these findings, additional sampling of *P. villosa* sympatric to the northern *P. maritima* subpopulations is recommended to conclusively determine if any distinction exists between *P. maritima* and *P. villosa*. Gene flow is evident among geographically close subpopulations of *P. maritima* in the north, particularly in Bare bluff, Woolgoolga headland, Pebbly beach, and Evans head sites. If the subpopulation of *P. maritima* in the north is tested to be genetically distinct, or if management actions will be deemed to be necessary to conserve the prostrate form, we can provide the necessary combinations of propagules to select to involve facilitate gene flow between subpopulations to increase diversity and reduce the risk of inbreeding depression.

Conservation genomics of *Rhodamnia rubescens* and *Rhodomyrtus psidioides* in support of management.

Stephanie Chen, Jia-Yee Samantha Yap, Jason Bragg and Maurizio Rossetto

The genetic diversity and structure of *Rhodamnia rubescens* and *Rhodomyrtus psidioides*, two rainforest Myrtaceae species critically endangered due to myrtle rust, were measured using high quality genome scans. We find both species' genetic diversity is spread along a latitudinal gradient and that current ex situ collections at the Australian Botanic Garden Mount Annan and Booderee Botanic Gardens adequately capture the species' genetic diversity across NSW. Further sampling north into QLD would likely be advantageous to capture unsampled genetic diversity. Kinship analyses revealed that clones are present in the ex situ collections; additionally, we inferred the source of individuals where it was unknown and consequently provide recommendations for efficient resource allocation in maintaining ex situ plants. As an example, we performed analyses to identify sets of individuals (of sizes N = 6 to 16) that maximize genetic diversity. This can guide the selection of individuals for use in cryopreservation and tissue culture. The maximization of genetic diversity will reduce the risk of inbreeding and increasing the adaptive potential to environmental change and other pressures. The genetic data and recommendations from this study will facilitate the on-ground management of *Rhodamnia* and *Rhodomyrtus* to contribute to their conservation management.



Other unpublished reports

Conservation genomics of *Caesia parviflora* var. *minor* in support of management and translocation activities.

Jia-Yee Samantha Yap and Maurizio Rossetto.

Prepared for RPS Australia Asia Pacific.

Taxonomic status and genetic health of the endangered *Caesia parviflora* var. *minor* was assessed using high quality genome scans. We provide empirical evidence confirming that the variety and *C. parviflora* var. *parviflora* are genetically similar and thus should be reassessed as belonging to one taxon. As our study only included a subset of both, broader sampling across the geographic distribution of both is recommended to determine the true geographic extent of this taxon. As for *C. parviflora* var. *minor* at Newnes, low genetic diversity is present, indicating that genetic rescue involving the mixing with nearby populations will be beneficial.



Gondwanan Rainforest Restoration Guidelines for Climate Change Adaption Strategy. Australian Institute of Botanical Science.

Richard Dimon, Jason Bragg, Marlien van der Merwe and Maurizio Rossetto.

We use the Restore and Renew knowledge infrastructure framework to guide the establishment of rainforest adaptive capacity, by providing detailed practical approaches on how to maximise evolutionary potential from evolutionary neighbourhoods (regionally local provenance) within protected areas. Overall, this report showcases how to reduce risk of local extinction of species and communities by providing practical, directly applicable guidance. We present planting guidelines based on targeted sourcing of a mix of seed/seedling material that is evolutionary diverse (adaptable) and climate resilient (adapted). We suggest a range of plant sourcing scenarios for dominant key stone rainforest species, including: maximising local diversity; supplementing local diversity from within genetic provenance; supplementing diversity from climate proofed provenance only; mixing climate-proofed and local diversities; and maximising diversity by enhancing with non-local provenances.

Conservation genomics of *Marsdenia longiloba* in support of management.

Elish McMaster, Maurizio Rossetto, Jia-Yee Samantha Yap, Julian Radford-Smith

Prepared for Sandpiper Ecological Surveys Pty Ltd

Marsdenia longiloba is an endangered species of milk vine found from Barrington Tops in New South Wales to Mount Nebo in Queensland. Part of its distribution is currently facing imminent removal due to the Coffs Harbour bypass project. This genomic study was requested to determine the extent of clonality in the affected subpopulations only, the proportion of diversity which will be impacted by the project, which individuals should be translocated to preserve local maximum genetic diversity, and if there are any trends in diversity that explain *M. longiloba* low reproductive output. Based on the SNP data of 177 samples, *M. longiloba* was found to be highly clonal at the Coffs Harbour sites, with only 87 genetic individuals found. Despite this, diversity remained adequate across the remaining clones. The trends in diversity indicate an outcross preference, with the high clonality and isolation of sites leading to the low reproductive output observed. Additionally, 7% of the total common alleles observed would be lost if all affected sites were wiped out. To decrease the effects of inbreeding depression and enhance reproductive output and to increase overall adaptive resilience we have determined optimal translocation populations of 20 and 35 individuals, which respectively capture >95% and >99% of the common allelic diversity observed.

Conservation genomics of *Pittosporum* sp. Coffs Harbour

Jia-Yee Samantha Yap and Maurizio Rossetto

Prepared for Transport NSW

A species-wide conservation genomic study of the newly discovered *Pittosporum* sp. Coffs Harbour was conducted to assess the genetic health, investigate the potential causes of its low reproductive output, and genomically select individuals to target for propagation in order to establish viable ex situ collections. Kinship analysis detected high relatedness among individuals from across the whole distribution. Diversity measures obtained across all populations detected reasonable levels of genetic diversity remaining across *P. sp.* Coffs Harbour. All sites were differentiated, suggesting that at least one representative from each site is required to preserve overall species' evolutionary potential. Optimised scenarios representing selection of ex situ individuals to be included in representative collections that capture diversity within impacted sites and across the whole species are presented.



Blog posts

Why is genetics relevant to ecological restoration?

Tricia Hogbin

Genetic information can help ensure restoration plantings are of appropriate provenance and sufficiently genetically diverse to be resilient in the long term. Fail to adequately consider genetic diversity when sourcing material for your restoration project and you may end up with populations that are unable to adapt to change and exhibit low or no recruitment.

Key points:

- Sourcing seed from a restricted geographic range or from one or few populations can decrease survival and recruitment of restored populations.
- Restored populations need genetic diversity to adapt to change and successfully reproduce.
- Low genetic diversity equals low adaptive potential.
- Given the extent of habitat loss and our changing climate, limiting ourselves to collecting only local provenance material may no longer be adequate, particularly when it comes to restoring threatened ecological communities..
- We need real live genetic data rather than generalisations.
- We can now easily gather a lot of relevant genomic information very quickly, very cost-effectively, making it feasible for restoration practitioners to access the information they need to design effective ecological restoration projects.

Restore and Renew – making genomic and climatic information freely available to restoration practitioners

Tricia Hogbin

The Restore & Renew project responds to the need for ecological restoration practitioners to incorporate the latest science into their toolkit, helping them to restore diverse, resilient and adaptable ecosystems. Resilient ecosystems need to be made up of species that are not only adapted to the local geology, climate and soil, but to future conditions as well. Restore & Renew acquires empirical knowledge on genetic diversity, habitat availability (current and future) and distributional patterns across multiple species to deliver restoration guidance to practitioners in easy to use publicly available web tools.

Key points:

- Restoration practitioners can use the Restore and Renew webtools to consider genetic information and climate change when sourcing material for ecological restoration projects and the establishment of seed production areas.
- A Site Matching Tool enables environmental matching of restoration sites to potential seed collection areas taking into account future climate change impacts. The tool can guide collection of seed that may be future-adapted and climate-ready.
- Practitioners can use the Restore and Renew Webtool to identify genetic provenance boundaries to guide collection of local provenance material.
- The Restore and Renew Webtool can help avoid unnecessarily restrictive seed collection due to underestimation of genetic provenance area.

Using Restore and Renew webtools to guide restoration of threatened ecological community Cumberland Plain Woodland

Tricia Hogbin

A new research project by the Research Centre for Ecosystem Resilience (ReCER) is exploring how genomic information can guide the restoration of threatened ecological communities and will help expand the application of the Restore and Renew webtools from a species-by-species approach to regional scale or community-specific restoration guidelines.

Key points

- The availability of suitable genetically diverse ‘climate ready’ material is a major constraint to the effective restoration of threatened ecological communities.
- Sampling from further afield and Seed Production Areas can increase available material—but should be guided by genetic data rather than generalisations to avoid negative consequences.
- A new project is using genomic information to guide seed sourcing for restoration of the threatened ecological community Cumberland Plain Woodland.
- Research outcomes will help expand application of the Restore and Renew webtools from a species-by-species approach to regional scale or community-specific restoration guidelines that will help practitioners maximise the self-sustainability, overall resilience and climate readiness of restored populations, while optimising return of investment.

AIBS Field Collection App launched

In 2022 ReCER launched the AIBS Field Collection App. This mobile app enables plant field collectors to accurately record field data associated with samples using a digital device (smart phones and tablets).

The app was designed as a data collecting tool for the Restore and Renew project and ensures consistent and reliable collection and management of metadata for the thousands of samples being collected across the state and beyond.

The app is freely available and can be downloaded from the Apple App Store or Google Play Store. More information at <https://recer.org.au/aibs-field-collection-app/>

