Genetic works to delineate threatened flora species for recovery priority setting – *Dryandra mimica* (***Banksia mimica***)

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Our environment, our future

Implementing priority actions for the recovery and management of nationally threatened EPBC Act listed species and ecological communities in Western Australia.

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Project 4: Genetic works to delineate threatened flora species for recovery priority setting – (1) *Dryandra mimica* (*Banksia mimica*)

Background

Dryandra mimica (now known as *Banksia mimica*) is ranked Vulnerable and is known from three population groups with a disjunct distribution. Populations occur at Mogumber near Gingin, north of Perth, and the Whicher Range near Busselton, south of Perth. Plants at Wattle Grove on private property are believed to have been destroyed through land clearing but a few plants were recently located in nearby Hartfield Park. Populations at Mogumber are quite large but the population at the Whicher Range has three patches of plants that may be clonal. *Dryandra mimica* has underground stems and is known to resprout after disturbance such as fire. If all populations represent the same taxon then the species would be recommended for removal from the WA Threatened Flora list. However this requires confirmation due to the disjunct distribution and knowledge of genetic differentiation in other plants with similar distributions.

Genetic analysis to determine the genetic structure between the population groups has been requested by the WA Threatened Species Scientific Committee. This project will develop suitable genetic markers for *Dryandra* and undertake genetic analysis to determine the pattern of differentiation between populations of the species at the three locations. This will indicate whether the species populations represent one species or whether taxonomic revision to recognise multiple taxa is required.

Activity

Undertake genetic analysis of *Dryandra mimica* to determine the genetic structure of the populations.

Results

Collections of leaf material were made from populations in each of the three locations as in Table 1. Plants in the Wattle Grove and Mogumber populations were discrete plants up to 0.5m in diameter and clearly separate from their neighbours. Plants in the Whicher Range population occurred in three patches, with large numbers of clusters of 5-15 leaves sprouting directly from the ground and separated by centimetres. The largest patch covered an area of approximately 10 x 15m. The form of the plants at Whicher Range suggested vegetative spread through underground stems and the presence of clonality within the patches. Potential clonal reproduction was also noted in the Clewleys Road population were there were some closely spaced neighbouring plants of differing sizes.

Samples were also collected from plants in two populations of the related species *D. vestita* across a similar geographic distance, at Mogumber and Brookton.

Microsatellite markers were developed for Dryandra through construction of a genomic library by Genetic Identification Services (USA). 20 primers were tested on 4 plants from each of the locations and 10 primers for selected for analysis of all plant samples.

DNA was extracted from all plants using Qiagen mini-kits. All plants were genotyped for the 10 microsatellite loci. Genotypes were scored from the raw data and analysis of genetic diversity and population structure was carried out using GenAlEx (v. 6).

Table 1. Details of populations sampled for genetic analysis

Location	Site	No. plants sampled	Comments
Whicher Range	Nature Reserve	20	Plants sampled from three patches
Wattle Grove	Hartfield Park -west	5	All plants sampled
Wattle Grove	Hartfield Park -east	5	All plants sampled
Mogumber	Clewleys Road	27	Large population
Mogumber	Fynes Road	20	Large population
D. vestita	Mogumber	20	
D. vestita	Brookton	20	

The microsatellite loci showed moderate levels of diversity with 100 alleles over the 10 loci and an average of 12.5 alleles per locus per populations. In the Whicher Range populations all samples within each patch had identical genotypes but samples from different patches had different genotypes. This indicates that each patch represents a single clonal genotype. Within the Clewleys Road population three sets of neighbouring samples (2, 2 and 5 samples) of differing sizes that could possibly be representative of clonal reproduction were sampled and these samples all showed the same genotypes indicating that clonal reproduction also occurs in the Mogumber populations.

The diversity within populations was moderate with observed heterozygosity ranging from 0.278 to 0.43 and expected heterozygosity ranging form 0278 to 0.455. Similar levels of diversity were seen in the two populations of *D. vestita*.

Within *D. mimica* there was a large amount of genetic differentiation with overall F_{ST} of 0.261 and 17% of the variation distributed between populations. The populations at Mogumber and Wattle Grove clustered together and showed the highest level of similarity. The population at Whicher range was genetically differentiated from the Mogumber and Wattle Grove populations. Differentiation between Whicher Range and the other populations accounted for the majority of the differentiation within the species. The Whicher Range population showed significant alleles frequency differences at 5 of the 10 loci and 53 of the 100 alleles were specific to either the Whicher Range or the Mogumber/Wattle Grove populations. A UPGMA diagram of the relationship between populations is shown in Figure 1.

The level of differentiation between the populations of *D. vestita* can be used as a guide to the level of differentiation expected between populations of a species over similar geographic distances. The level of differentiation in *D. vestita* was similar to that between the Mogumber and Wattle Grove populations of *D. mimica*. (F_{ST} 0.15, 18 specific alleles in D. vestita; F_{ST} 0.10, 10 specific alleles in Mogumber/Wattle Grove). In comparison the Whicher Range populations showed a much greater level differentiation from the Mogumber/Wattle Grove populations (F_{ST} 0.29, 53 specific alleles). A Principal Co-ordinates Analysis of the relationships between species is shown in Figure 2.

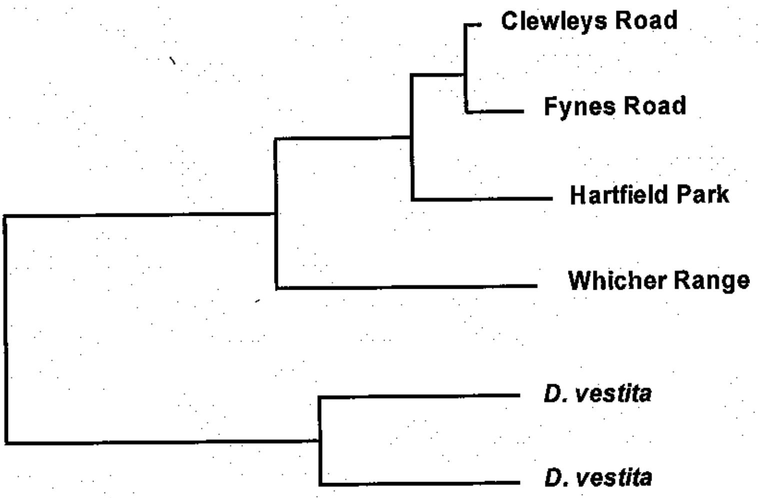

Figure 1. The relationships between populations of *D. mimica* and *D. vestita*

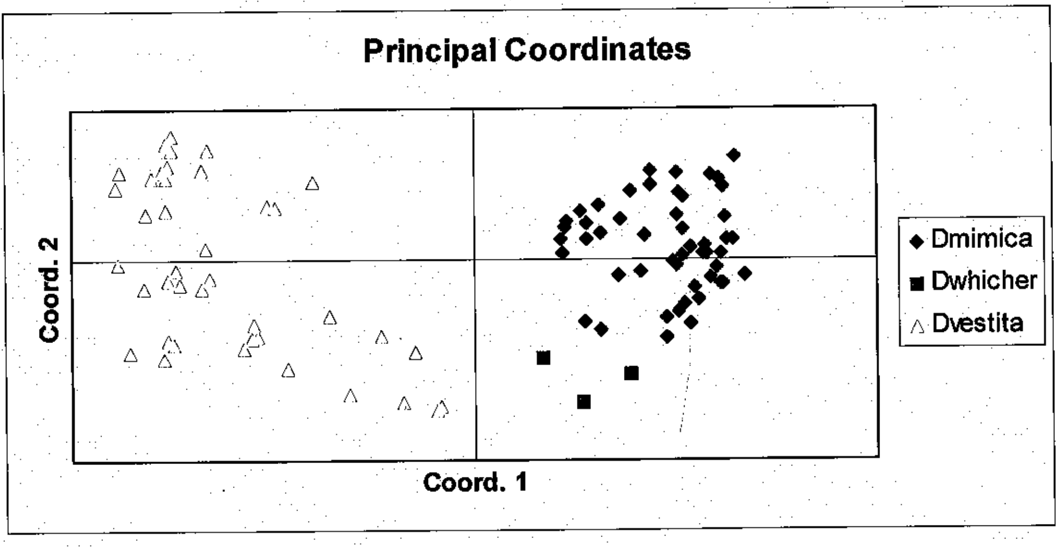

Figure 2. PCA analysis of the relationships between taxa.

Outcome

The level of genetic divergence between the Whicher population and the Mogumber/Wattle Grove populations indicates that these populations represent separate taxa. Therefore a taxonomic revision is required and further assessment of the conservation status of the two taxa should be undertaken. The small size of the Whicher Range population suggests it would be ranked with a high conservation status although it does occur in a Conservation Reserve so is not under immediate threat and its clonal reproduction indicates a capacity to resprout.