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SJSU Undergraduate Research Grants

Population Genetics of *Pedicularis dudleyi*, Redwood Forest Specialist

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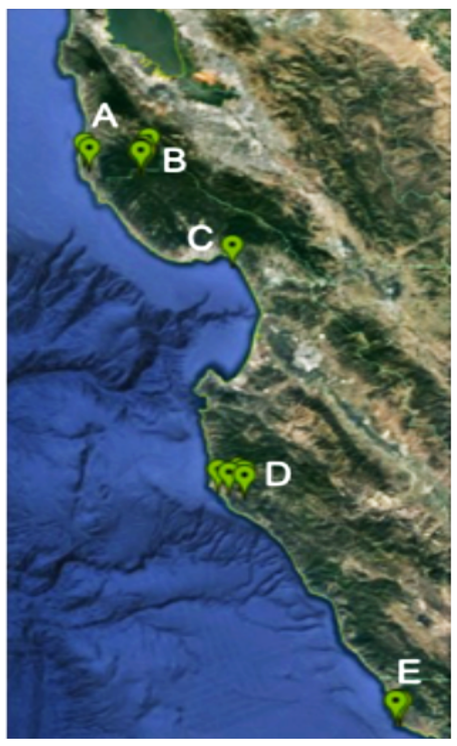
Abstract

Pedicularis dudleyi, commonly known as Dudley's Lousewort (**Fig. 1**), is a wildflower endemic to the redwood forests of central California. Locality data from herbarium specimens suggest that the species is distributed across four or five populations along the coastal regions of California. However, the extent to which these populations are genetically isolated from each other is unknown. Furthermore, morphological differences observed in individuals from population E (**Fig. 2**) have led some experts to suggest *P. dudleyi* may represent two species. This work represents the first population genomic study of *P. dudleyi* and aims to :

1) **Quantify the genetic diversity within populations of *P. dudleyi***
2) **Infer the extent of gene flow among populations**
3) **Use molecular data to test the hypothesis that *P. dudleyi* represents two distinct species.**

Fig. 1: *P. dudleyi* found in Portola Redwood State Park, or population B. **Fig 2**. Right shows all populations of *P. dudleyi* along coast of California. Sampling for this study included individuals from populations A, B and E.



Methods

Samples of *P. dudleyi* were collected from populations at Pescadero Marsh and Portola Redwood State Park (population A and B in **Fig. 2**)

Double Digest Restriction Site-Associated DNA sequencing (ddRAD-seq)(Fig. 3 A-C): ddRAD-seq works by partitioning genomic DNA into smaller fragments using enzyme pairs that recognize specific restriction sites and cleave the DNA at those positions. Fragmented DNA is then enriched for library preparation and sequenced using next generation sequencing technology. In this manner it is possible to obtain hundreds and sometimes thousands of single nucleotide polymorphism (SNP) markers from across the entire genome for robust population genomic analysis. DNA fragments ranging from 600 -700bp and sequenced on one lane of an Illumina HiSeq4000 (100 bp, single-end) at the QB3 Vincent J. Coates Genomics Sequencing Laboratory at the University of California, Berkeley.

Fig. 3A. We extracted DNA from leaf material using a Qiagen DNEasy Mini Plant kits.

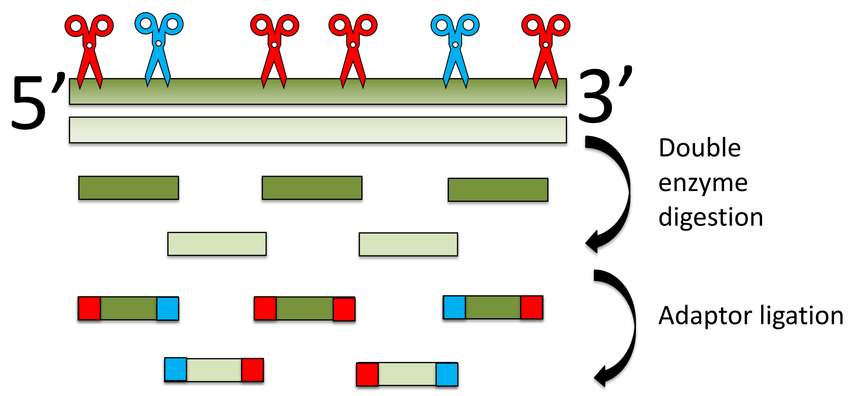


Fig. 3B. DNA was digested using the enzyme combination *EcoRI*+ *SphI* which were tested *insilico*. Custom barcodes and adapters were ligated to each sample.

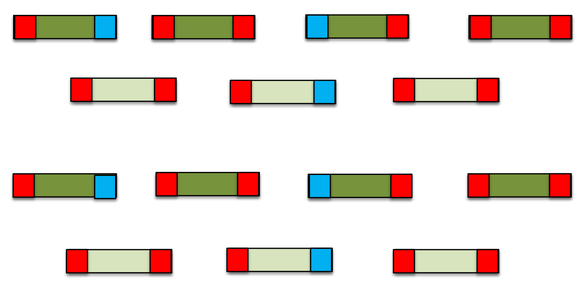


Fig. 3C. Samples were pooled and then amplified using PCR to produce Illumina sequencing libraries.

Next Steps

In order to understand how populations are genetically structured and the extent to which gene flow is occurring between them, we will use SNP data to calculate summary statistics such as observed heterozygosity, nucleotide diversity, and F-statistics and to conduct Bayesian clustering analysis implemented in STRUCTURE (Pritchard et al. 2000). Understanding the genetic structure of *P. dudlyei* will aid in our end goal of helping public land managers in developing strategies to maintain the health and genetic integrity of Dudley's Lousewort.

Citations

- Consortium of California Herbaria database. http://ucjeps.Berkeley.edu/consortium, accessed Sept. 10, 2018.
- Vorobik, L. A. 2012, Pedicularis dudleyi, Jepson Flora Proect (eds.) Jepson eFlora,
- Yang, G.Q., *et a*l. 2016. Development of a universal and simplified ddRAD library preparation approach for SNP discovery and genotyping in angiosperm plants. *Plant methods*, *12*(1), p.39.
- Pritchard, J.K., Stephens, M. and Donnelly, P., 2000. Inference of population structure using multilocus genotype data. *Genetics*, 666*155*(2), pp.945-959.6