

Variation of fungi in *Corallorhiza maculata* between distinct populations in the northwest and southwest of the United States

Introduction

Corallorhiza maculata is a terrestrial mycoheterotrophic orchid. Myco-heterotrophs are nonphotosynthetic plants that consume fungi. The orchid does not do anything beneficial for the fungus. The fungus is persuaded to spread and increase within the mycorrhizal organs of the orchid (1). In a normal plant's relationship with mycorrhizal fungi, the plant supplies sugars. In orchids it digests the hyphae of the fungi that colonize them (2). The majority of plants keep their ectomycorrhizal fungi on the outside of the root, however this same fungus lives inside of orchid roots also known as rhizomes. Previous studies have shown that C. maculata affiliate only with ectomycorrhizal fungi within a single basidiomycete family, the Russulaceae (2).

In this study, the samples originate from two geographically distinct areas, the Northwest (north California, Oregon, Washington) and Southwest (Arizona, New Mexico) of the United States. The main objective of this project is to find out if the fungi in two populations of orchids make-up two distinct clades or intermingle in one clade. We are interested in knowing if the Rocky Mountains (geographical barrier) are a barrier to a gene flow. If there is a barrier, we are figuring out if it is affecting the fungi that the orchid will associate with. We hypothesize the orchids aren't mating between the two populations and eventually evolutionarily diverging.

Methods and Materials

On 6/10/08, we collected *Corallorhiza trifada*. We extracted DNA from our Ohio samples and two of my samples (*C. trifada*). We ran a PCR using primers ITS 1F and ITS 4 (forward and reverse). Next, we performed PCR clean up. Then extracted genomic DNA from seven additional samples. Then we amplified our DNA via PCR. We used a Nanodropp to see how much DNA was extracted. We submitted the 16 samples to the UAF DNA core lab and got our sequences. Then we cleaned our reads in Sequencer, and used Bioedit to build a sequence alignment. To finish we built a phylogenetic tree using the program MEGA, which included the GenBank sequences used to compare with our DNA samples.

Daphne Attla, Sarah Hopkins, Dr. D. Lee Taylor CFB 40 bR Russula cf. cyanoxantha CFB 40 aR Russula laricina Russula laricina Gymnomyces monticola JVF 2833 I Russula curtipes Gymnomyces abietis Russula risigallina CFB 34 aR CFB 50 aR - Russula turci 86 1 C maculata Mt San Jacinto CA Russula queletii – Russula fragilis var. fragilis CFB 31 bR 14 5 C maculata Type O r Russula aff. delica JVF 2826 I Key Northwest= Blue Southwest= Red 0.01

Results/ Discussion

- •As a result of my research, I found there is no apparent distinction in the ITS locus between the N.W. and S.W. orchid fungi. The reason may be that we only sampled one variety of *C. maculata. We* found the orchid fungi to be in one clade.
- •The total tree length for all samples (excluding the GenBank sequences): 289 base pair changes
- •The southwest tree length: 164 bp. changes. We expect this because the orchids have inhabited in the southwestern region of the U.S. for a longer period compared to the N.W. Therefore these orchids have been able to diversify because they are able to use a bigger variety of fungi.

The northwest tree length: 106 bp. changes. The northwest samples weren't able to diversify as much as the southwest samples because it hasn't been in that region as long.

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Literature Cited

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- (2) Taylor, D. L., and T. D. Bruns. "Independent, Specialized Invasions of Ectomycorrhizal Mutualism by Two Nonphotosynthetic Orchids." <u>Proceedings of the National Academy of Sciences of the United States of</u> 94.9 (1997): 4510-5.

