ON THE EVOLUTION AND PHYLOGEOGRAPHY OF THE SOUTHEASTERN SPECIES OF THE GENUS DALEA L. (FABACEAE) USING A PHYLOGENETIC APPROACH

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Acknowledgement

We would like to thank the University of North Georgia- Gainesville Campus Department of Biology for allowing us use of lab equipment and providing funding for our research. We would also like to thank the University of North Georgia's CURCA program for providing us with additional grant monies to also help fund our research.

Introduction

The fields of phylogenetics and phylogeography focuses on the relatedness of species through temporal and spatial lenses. (Avise 2000) These fields grow every year as science's understanding of the genetic code continues to expand, and this research study will contribute further to these studies with a focus on botany by reviewing how a genus of southeastern native plants, *Dalea* (Fabaceae) (Fig. 1 &2),


(Fig. 1: *Dalea pinnata var. trifoliata*)


(Fig. 2: *Dalea mountjoyae*)

diverged and capitalized on certain geographical niches. The genus *Dalea* consists of approximately 160 species with a few species being geographically widespread, and most of the genus being endemic to small restricted areas making it an indicator species for habitat loss. (Barneby 1977) A previous study looked at the phylogeny of the tribe which *Dalea* belongs to, Amorpheae, and only one other phylogenetic study has been conducted on the genus but focuses on species endemic to Alabama and Georgia. (Diggs 2013, McMahon and Hufford 2004) This study focuses on *Dalea* species from the Gulf Coastal Plain of Florida, including *D. carnea*, *D. carthagenesis var. floridana*, *D. feayi*, *D. pinnata var. pinnata*, *D. pinnata var. trifoliata*, *D. adenopoda*, *D. mountjoyae*, and *D. albida*.

Methods & Materials

Tissue samples of *D. carnea*, *D. carthagenesis var. floridana*, *D. feayi*, *D. pinnata var. pinnata*, *D. pinnata var. trifoliata*, *D. adenopoda*, *D. mountjoyae*, and *D. albida* were collected in the field the summer of 2016, and locality of each individual specimen were noted. DNA was extracted (Fig. 3)


(Fig. 3: Research Team Performing CTAB DNA Extraction)

from the tissue sample of *D. carnea*, *D. carthagenesis var. floridana*, *D. feayi*, *D. pinnata var. pinnata*,and *D. adenopoda* using a standard CTAB extraction protocol but with a modified CTAB with the addition of 0.18g Spermine, 0.13g Spermidine, and 200ml BME. (Diggs 2013, Doyle and Doyle 1987) PCR amplification has been run on all extracted samples using the chloroplast gene primers *matK-xf/-malp* and a PCR profile of: 4 min at 94°C, 35 cycles of 1 min at 94°C, 1 min at 44°– 48°C, and 3 min at 72°C, followed by a final extension of 5 min at 72°C. (Kusmina 2013, McMahon and Hufford 2004) DNA amplification products will be sent to a genomic laboratory for gene sequencing, and phylogenetic trees will be constructed using various statistical methods.

Literature Cited

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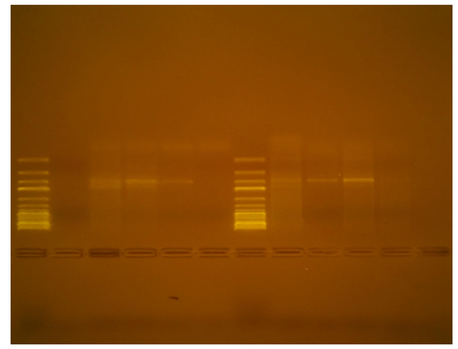
Results

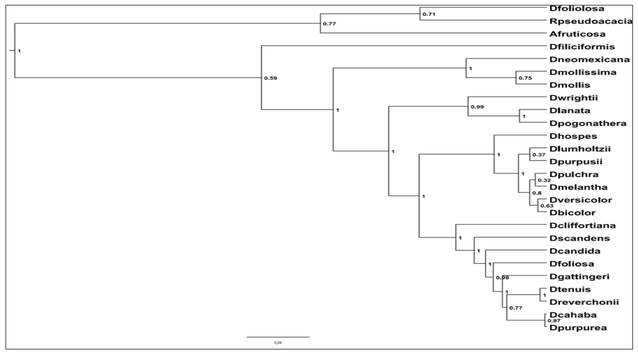
All target species have been collected, and DNA extractions have been preformed on the following species: *D. carnea*, *D. carthagenesis var. floridana*, *D. feayi*, *D. pinnata var. pinnata*,and *D. adenopoda*. DNA extractions from tissue samples of *Dalea* have yielded low DNA concentrations averages, but UV absorbance ratios have shown fairly pure DNA solutions. PCR amplification produced positive results with the first samples trialed (Fig. 4),


(Fig. 4: Gel Electrophoresis Showing Positive Bands)

but these positives have been lost with later samples. Further results will be analyzed as progress continues.

Discussion

PCR amplification can be challenging for the Genus *Dalea* due to high concentrations of volatile organic compounds, and aromatic protein complexes that bind to DNA. New DNA extraction and PCR procedures are currently being developed by our research team. Based on previous studies (Fig. 5), we suspect that there will be three distinct lineages of *Dalea*, one from a Texas origin, one from Gulf Coastal Plains origin, and an outlier lineage with *D. carthagenesis var. floridana* which origin remains highly speculative. (Diggs 2013)



(Fig. 5: Phylogenetic Tree Of The Genus *Dalea*, With Branch Length Based On Proportions Of Time)