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

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INTRODUCTION
The fields of phylogenetics and phylogeography focus 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).

METHODS & MATERIALS
Tissue samples of D. carnea, D. carthagenesis var. floridana, D. feayi, D. pinnata var. pinnata, D. pinnata var. trifoliata, D. adenopoda, D. montjoiyae, 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)

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 48°– 48°C, and 3 min at 72°C, followed by a final extension of 5 min at 72°C. (Kuzmina 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.

RESULTS
All target species have been collected, and DNA extractions have been performed 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), 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)

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LITERATURE CITED

On the evolution and phylogeography of the southeastern species of the genus Dalea L. (Fabaceae) using a phylogenetic approach.