Does *Pinus edulis* genetic diversity explain increased susceptibility to drought-associated mortality?

BACKGROUND

Climate change increases drought-associated tree mortality,[1] converting forests from carbon sinks to carbon sources[2] and undermining the role forests play in regulating climate.[3] Understanding and predicting widespread mortality is critical because extensive die-off events influence local and regional climate and vegetation.[4] Recent efforts have focused on improving mortality thresholds by focusing on climate variables, physiological responses, and multi-step calculation metrics that combine climate, eco-hydrological and ecophysiological data.[5] These metrics are critical for understanding rates of mortality but offer little insight into adaptive strategies that may be used to mitigate drought-associated mortality such as conservation genetics. For example, subpopulations may be locally adapted to particular environmental conditions and may be maladapted when exposed to new conditions. **To overcome this limitation, I will characterize the regional genetic structure of piñon pine and compare haplotype diversity with population level mortality rates to determine the role that genetics has in influencing the likelihood of piñon pine mortality.**

Some evidence suggests that genetics may play a larger role in drought-associated mortality than previously acknowledged. A study of piñon pine mortality during drought found a trade-off between resistance to the moth *Dioryctria albovittella* and increased susceptibility, up to three times higher, to mortality.[6] Additionally, Gehring et al. 2017 found that ectomycorrhizal community composition is under strong genetic control and that these distinct EMF communities differentially impacted piñon pine trees' survival and growth.[7] The most direct evidence of piñon pines genetic control was a study done on climate mediated geographical adaptive differences for a Gly polymorphism.[8] This population genomic approach found that different Gly genotype

exhibited fitness differences and that Gly-33 homozygotes were only found in sites with low precipitation. These three studies are examples of piñon pines' genetic explanatory power and the potential it may have in understanding the large intra-specific variation in drought-associated mortality.

I hypothesize (**H1**) that areas with high levels of piñon pine genetic diversity will have lower levels of drought-associated mortality. During the last glacial maximum, piñon pine (*Pinus edulis)* trees' population was fragmented into two distinct refugia.[9] An ancestral refugium in southern New Mexico and northern Texas and a younger refugium in central Arizona. Post glacial retreat, piñon pine trees migrated northward as abiotic conditions permitted to form extant population range. These two genetically different glacial refugial populations should have substantial differentiation among them. Alternatively, I hypothesize that (**H2**) the large-scale morality events of the last century[1] severely reduced the genetic diversity variation among populations and may obstruct the diversity-mortality relationship. Support for **H2** would still be invaluable because it would characterize piñon pines population structure, which is critical to know as more frequent and hotter droughts are expected to increase in the southwestern US.
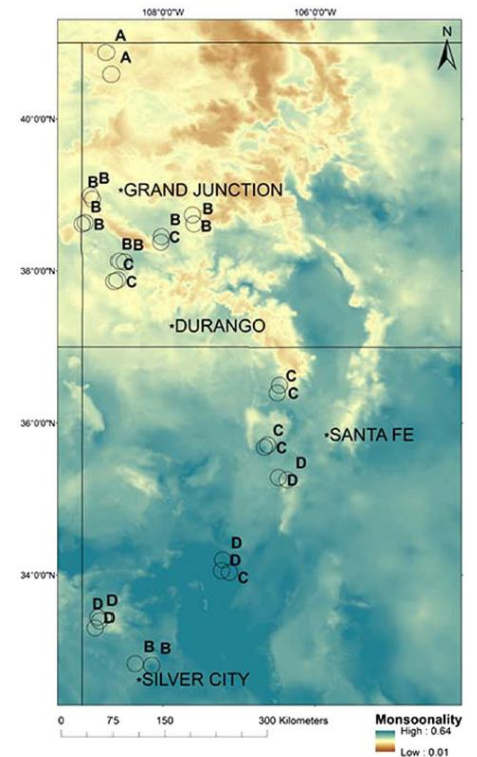
METHODS

In Fall 2020 I will visit 28 sites that span an elevation (1000 m) and latitude (1100 km) gradient (Fig 1). These sites were identified and established by Wion et al. 2019 using existing vegetation classification system. Sites span an elevation from 1480 to 2420 m and span a mean annual precipitation from 300m to 630 mm. At each site I will choose 3 piñon pine trees at established 50-m transects and will take needle samples of individual trees (N =84). Needles will be stored in silica until I extract DNA (Qiagen DNeasy plant mini kit). Three regions of mitochondrial

(mtDNA) and nine regions of chloroplast DNA (cpDNA) will be surveyed. Both mtDNA and cpDNA will be measured because the former is maternally inherited, and the latter is paternally inherited. Mortality data will be provided from collaborators. In instances where mortality data is missing the Forest Inventory and Analysis (FIA) morality surveys will be utilized to infer regional rates. Additional data from these plots that were already collected include tree growth, seed cone production, height, and canopy.

Figure 1: Field sites from Wion et al. 2019 Background color shows moonsoonality gradient. Moonsoonality is a measurement of the average seasonal water deficit

SIGNIFICANCE

The Desert Southwest is one of the most sensitive ecoregions and overlaps with a continental climate change hotspot.[10] Understanding the role of genetic diversity in predicting drought-associated mortality will enhance our ability to develop adaptive management strategies such as translocation and aid conservation genetic efforts. This is particularly important because, piñon-juniper woodlands span over 24 million ha and are the third largest biome in the United States. They contain relatively high levels of biomass and reliably sequester carbon. Finally, the completion of this study will serve as my third chapter for Ph.D. dissertation and the expected results of this work will be presented at the 2021 Ecological Society of America meeting.

BUDGET:

Item	Description	Cost per unit	Total needed	Total cost	Funding source
Fieldwork	Travel to sampling sites ≈ 613 miles to cluster A (Northern CO) ≈ 476 miles to cluster B (Southern NM)	$0.78 /mile	1089 miles	$849.42	Lewis & Clark *received
Qiagen DNeasy plant mini kit (50 samples/kit)	DNA extraction kit for plants	$243	2	$486	**GRAC ($400)** UNM GPSA SRG *pending
Total = $1,335.42			**Total GRAC requested = $400** Total received from others = Total requested from others = $355.72		

Animal Care and Use: Not needed. No animals are involved in this proposal.

Previous use of GRAC funds:
I received GRAC funds in spring 2019 (*Does past performance predict future survival in Pinus edulis*) and 2020 (*What makes a super producer? Testing the trade-off between reproduction and growth in masting species*). 2019 fund were used to pay for carbon isotope analysis of tree rings. Four different trees were analyzed at an annual resolution. Two of the trees had survived the recent drought of 2013 and the other two trees were drought-killed pines. I found that the drought-killed pines demonstrated greater sensitivity in growth and larger variation in carbon isotope values many years prior to the 2013 drought indicating early on susceptibility. The results from this study were presented at the Ecology Society of America's conference in 2019. 2020 funds were used to visit field sites at the Sevilleta National Wildlife Refuge and to cover dendrochronology equipment costs. 50 trees were cored, cross-dated, and ring widths were measured. I found a weak relationship between cone production and basal area growth. I had hypothesized that piñon pines with large cone production (high mast seeding) would have lower basal growth and that piñon pines with low minimum cone production (low mast seeding) would have higher basal growth following a resource allocation trade-off. I did not find support for differences in resource allocation between high and low mast seeding pines. Despite different cone production there was a high degree of synchrony across the trees which suggests that the pines are cueing in on a common weather signal and must have a baseline level of resources for cone production. These results were presented in at the Ecology Society of America's conference in 2020.

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