

## **Patterns of Whitebark Pine Carbon Allocation: Implications for Stress Resistance and Conservation Strategies**

### A. Background and Objectives

Whitebark pine (*Pinus albicaulis*) is a high-elevation pine species found throughout the North American Rocky Mountains. It is considered a keystone and foundational species, due to its large, nutritious seeds; its ability to establish in exposed, cold, windy sites, and the protection from these harsh conditions it provides for other species (Callaway, 1998; Maher et al., 2005). Whitebark pine also stabilizes snowpack, delays snowmelt, and prevents soil erosion (Schmidt and McDonald, 1990; Tomback et al., 2001). The combination of these traits promotes community biodiversity by creating locally stable conditions, affording habitat and nesting sites to a diverse array of species, and providing forage for wildlife (Tomback and Kendall, 2001). These attributes ensure that vectors of stress negatively affecting *P. albicaulis* populations can have far reaching effects on ecosystem health and function.

These trees are currently facing grave threats as a result of global change. Indeed, the species has recently been listed as “threatened” under the Endangered Species Act (USFWS, 2020). Modalities of risk include white pine blister rust infections (via an introduced fungus: *Cronartium ribicola*), outbreaks of mountain pine beetle (*Dendroctonus ponderosae*), and climate change, which may exacerbate the effects of the first two (Keane et al., 2012). The combination of these factors has resulted in a marked decline in whitebark pine populations, which could be accelerated by future warming trends (Hansen et al., 2016).

A primary conservation strategy put forth for mitigating these threats is restoring whitebark pine stands through the outplanting of seedlings (Keane et al., 2012). It is essential to this strategy that the seedlings chosen for restoration efforts are able to withstand the pressures they will face on the path to maturity. Much effort has been invested in tree improvement through identifying and propagating genotypes that exhibit resistance to white pine blister rust (Sniezko et al., 2018; Mahalovich & Dickerson, 2004). However, to ensure the future survival of whitebark pines, similar efforts must be made to identify and propagate genotypes exhibiting resistance to the harmful effects of climate change.

Mechanisms of climate-induced forest mortality include hydraulic failure, carbon starvation, and increased vulnerability to insects and pathogens (McDowell, 2011; Zeppel et al., 2011). However, further research is needed to understand and predict the impacts of climate change-induced stress on high elevation pine physiology and future forest dynamics. An important area of research that may illuminate how *P. albicaulis* will respond to rapid climate change, is patterns of seedling carbon allocation. By studying how individuals allocate carbon under climate change-type stress, we may gain valuable insights into patterns associated with high degrees of stress resistance and resilience to the different mechanisms of climate-induced mortality.

A key trait that confers stress resistance and resilience to trees, is the storage of nonstructural carbohydrates (NSCs). Storage pools of NSCs enable trees to maintain and recover hydraulic and metabolic functions both during and following periods of extreme stress, such as droughts and freezing temperatures. (Hartmann & Trumbore, 2016; Sevanto et al., 2014). While these NSC storage pools are essential for a seedling’s survival, it is yet unclear what mechanisms

affect variation in storage patterns, and the degree to which phenotypic plasticity and genetic adaptation play a role in controlling storage. By examining the variation in NSC storage and allocation between whitebark pine populations and individuals under stress, we may be able to predict species' ability to tolerate or adaptively evolve in response to rapid environmental change. Additionally, NSC patterns associated with high degrees of tolerance may inform restoration and conservation strategies.

To address these matters, we plan to experimentally impose heat, drought and cold stress treatments on seedling populations of *P. albicaulis* from contrasting climates. Under these conditions, we will quantify patterns of seedling carbon allocation in response to stress. We will then compare these stress responses among populations, and identify physiological markers associated with high degrees of stress resistance and resilience. Understanding these mechanisms will improve predictions of future vegetation dynamics and inform mitigation strategies.

### B: Study Plan

To conduct this research, we have established 2- and 3-year-old whitebark pine seedlings (donated from the USFS and Coeur d'Alene Nursery) in the Montana State University Plant Growth Center. We will utilize growth chambers to impose the heat, drought, and cold stress treatments on *P. albicaulis* populations. The cold stress treatments are being imposed during the winter of 2020-21. All other stress treatments will be imposed during the growing season of 2021. An array of metrics will be collected throughout the stress treatments including bud phenology, growth, gas exchange, and hydraulic functioning. Additionally, we will collect samples from all treatments, and all tissues (needles, branches, stems and roots) to measure NSC content throughout the duration of the experiments. We will measure both total NSC content, as well as patterns of allocation to different tissues.

### C: Measures of Success

The persistence of future whitebark pine ecosystems is predicated upon effective management and restoration strategies. Effective strategies in turn rely upon comprehensive scientific understanding. This study will improve our understanding of seedling responses to stress, and detect markers associated with high degrees of resistance and resilience. These findings will have the effect of informing tree improvement and outplanting strategies, thus increasing the likelihood of long-term success of restoration efforts. Consequently, due to the keystone nature of whitebark pine, this study has the potential to have cascading effects on future ecosystem health and function.

### D: Allocation of Funds

I am requesting the WPEF Student Grant to fund a critical part of my M.S. thesis research: the costs of NSC reagents and consumables. The NSC analysis process requires a costly array of reagents, and this grant would help offset the associated expenses, allowing a more detailed analysis of NSC allocation patterns. By increasing the resolution of the analyses we are able to complete, we may gain insight into patterns that would otherwise be indiscernible, therefore bringing us further on our path towards improved conservation strategies.

## References

- Callaway, R. M. 1998. Competition and facilitation on elevation gradients in subalpine forests of the northern Rocky Mountains, USA. *Oikos*, **82**, 561–573.
- Hansen, A., Ireland, K., Legg, K., Keane, R., Barge, E., Jenkins, M., Pillet, M. 2016. Complex challenges of maintaining whitebark pine in Greater Yellowstone under climate change: A call for innovative research, management, and policy approaches. *Forests*, **7**, 54.
- Keane, Robert E., D. F. Tomback, C. A. Aubry, A. D. Bower, E. M. Campbell, C. L. Cripps, M. B. Jenkins, et al. 2012. “A Range-Wide Restoration Strategy for Whitebark Pine (*Pinus albicaulis*).” RMRS-GTR-279. Ft. Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.
- Maher, E. L., Germino, M. J. & Hasselquist, N. J. 2005. Interactive effects of tree and herb cover on survivorship, physiology, and microclimate of conifer seedlings at the alpine tree-line ecotone. *Can. J. For. Res*, **35**, 567–574.
- Mahalovich M. F., Dickerson G. A. 2004. Whitebark pine genetic restoration program for the Intermountain West (United States), in Proceeding IUFRO Working Party 2.02.15 Breeding and Genetic Resources of Five-Needle Pines: Growth, Adaptability, and Pest Resistance, 23–27, July 2001. (Medford, OR; Fort Collins, CO: USDA Forest Service, Rocky Mountain Research Station; Proceedings RMRS-P-32, 181-187).
- McDowell, N.G., 2011. Mechanisms Linking Drought, Hydraulics, Carbon Metabolism, and Vegetation Mortality. *Plant Physiology*, **155**, 1051–1059.
- Schmidt, W. C. & McDonald, K. J. 1990. *Proceedings--Symposium on Whitebark Pine Ecosystems: Ecology and Management of a High-Mountain Resource, Bozeman, MT, March 29-31, 1989*. vol. 270 (US Department of Agriculture, Forest Service, Intermountain Research Station).
- Snieszko, R. A. *et al.* (2018) ‘Blister rust resistance in whitebark pine (*Pinus albicaulis*) - early results following artificial inoculation of seedlings from Oregon, Washington, Idaho, Montana, California, and British Columbia seed sources’, In: *Schoettle, Anna W.; Snieszko, Richard A.; Kliejunas, John T., eds. Proceedings of the IUFRO joint conference: Genetics of five-needle pines, rusts of forest trees, and Strobosphere; 2014 June 15-20; Fort Collins, CO. Proc. RMRS-P-76. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. p. 129-135., 76, pp. 129–135.*
- Tomback, D. F.; Achuff, P. 2010. Blister rust and western forest biodiversity: ecology, values and outlook for white pines. *Forest Pathology*. **40**, 186-225
- Tomback, D. F., Arno, S. F. & Keane, R. E. 2001. *Whitebark pine communities: ecology and restoration*. Island Press. Washington, DC, USA.
- Tomback, D. F. and Kendall, K. C. (2001) *Biodiversity losses: The downward spiral*. Island Press. Washington, DC, USA.
- United States Fish and Wildlife Service. 2020. Endangered and Threatened Wildlife and Plants; Threatened Species Status for *Pinus albicaulis* (Whitebark Pine) with Section 4(d) Rule. *Federal Register*, **85(232)**, 77408-77424
- Zeppel, M., Adams, H., & Anderegg, W. 2011. Mechanistic causes of tree drought mortality: Recent results, unresolved questions and future research needs. *The New Phytologist*, **192(4)**, 800-803.