

**AGRICULTURAL RESEARCH FOUNDATION
FINAL REPORT
FUNDING CYCLE 2019 – 2021**

TITLE: Population genomics of *Leptographium wagneri*: The importance of local versus long-distance dispersal in black stain root disease epidemiology

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EXECUTIVE SUMMARY: Beginning in the spring and summer of 2015 unusual mortality was observed in young (< 6 years old) Douglas-fir (*Pseudotsuga menziesii*) plantations in the central and southern Coast Range of Oregon. Affected trees had yellow to red crowns and were randomly distributed throughout the stand. Careful examination of the symptomatic trees revealed the presence of a black discoloration or staining below the bark. This combination of symptoms is characteristic of black-stain root disease (BSRD) caused by the fungal pathogen *Leptographium wagneri*. This fungus can spread from infected trees to healthy trees via root grafts, but is also known to be vectored by root feeding insects. Historically, BSRD is most damaging in 10–25-year-old stands that have undergone pre-commercial thinning (PCT) and in stands with stressed trees associated with roadsides, skid roads, and compacted soils. The incidence and pattern of mortality in stands younger than 10 years is alarming, has not been noted in the past, and cannot be attributed to PCT or soil compaction. Previous studies examining the epidemiology of BSRD have focused on trapping and identifying the insects that spread this disease. Determining which insects are actually infested with the fungus and how far those insects have dispersed poses significant difficulties. Sampling the pathogen directly and using molecular markers and population genetics to infer dispersal patterns can circumvent these difficulties. This approach has been extremely effective in determining how introduced and native pathogens spread across the landscape.

OBJECTIVES: The overall objectives were to develop a better understanding of the epidemiology of BSRD by using population genomics, provide recommendations to land managers that will aid in the development of effective management strategies for this disease, and to build a foundation upon which future studies of BSRD dynamics would be based. The specific objectives were: 1) Sequence the genomes of isolates of *L. wagneri* var. *pseudotsugae* to identify single nucleotide polymorphisms (SNPs) that vary within and between populations; 2) Compare genetic diversity and population structure within and between forest stands and plantations using SNPs; 3) Infer the relative importance of local vs. long distance dispersal in the establishment of new BSRD infection centers; and 4) Determine whether BSRD infections in newly planted stands are the result of infection from the previous stand, or are established independently via insect vectors.

PROCEDURES: Wood samples were collected from infected Douglas-fir trees in four Douglas-fir plantations. The BSRD fungus (*L. wagneri*) was isolated in pure culture from each infected wood sample. DNA extraction and Illumina sequencing were performed by the Center for Genome Research and Biocomputing (CGRB) at Oregon State University. Sequencing of the genomes and subsequent bioinformatic and population genomic analyses provided an estimate of the genetic variation among isolates. Analysis of the spatial distribution of genetic variation on the landscape allowed for inference of the reproductive biology, genetic diversity, and population structure of *L. wagneri*. This information was used to infer the mechanisms of pathogen dispersal and epidemiology of this disease.

SIGNIFICANT ACCOMPLISHMENTS: We sampled *L. wagneri* from ~ 160 Douglas-fir trees in five tree plantations spanning much of the Oregon Coast Range. The isolates have all been sequenced and the results have been analyzed. We are currently finalizing the publication reporting on the results and anticipate that it will be submitted by the end of summer 2021.

BENEFITS & IMPACT: The results of this research provides valuable insights into the ecology and epidemiology of a native insect-vectored vascular wilt pathogen that represents a major management concern in Douglas-fir plantation forestry. This study will provide information to land managers that could lead to the development of more effective management strategies for mitigating the impacts of black stain root disease. A manuscript summarizing these results is currently being prepared and will be submitted to Forest Ecology and Management.

ADDITIONAL FUNDING RECEIVED DURING PROJECT TERM: Weyerhaeuser Timber Co. gifted the OSU foundation and my research program an additional \$20,000 USD to support sequencing costs. Additional funding was also provided by USDA Forest Service Region 6 Forest Health Protection and Forest Health Monitoring EM Grant WC-EM-17-05.

FUTURE FUNDING POSSIBILITIES: Based on the results from this project, we intend to pursue additional funding to continue research. We are considering submitting proposals to NSF or the USDA-AFRI.