

Black Stain Root Disease Studies on Ponderosa Pine – Parameters and Disturbance Treatments Affecting Infection and Mortality

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INTRODUCTION

Black stain root disease of ponderosa pine (*Pinus ponderosa* Doug. Ex Laws.), caused by *Leptographium wageneri* var. *ponderosum* (Harrington & Cobb) Harrington & Cobb, is increasing on many eastside Sierra Nevada pine stands in northeastern California. The disease is spread from tree to tree via root contacts and grafts but overland spread of the disease is most likely due to woody root feeding bark beetle (*Coleoptera:Scolytidae*) vectors. Soil and site relations along with disturbance are factors in the etiology of the disease (Harrington and Cobb 1988). Thinning and prescribed burning are important silvicultural tools in maintaining forest health in eastside pine stands. Because soil compaction is a concern in many sites, skid trails are treated by subsoiling equipment to alleviate compaction where this might be an issue. However, little is known of the effects of these silvicultural treatments on incidence of black stain root disease on sites with high disease risk. Because the woody root feeding insects that vector the disease respond to disturbance (Otrosina – Ferrell 1995), understanding consequences of different disturbances resulting from silvicultural treatments is essential for devising management plans to mitigate disease impact. This paper summarizes preliminary results from two long-term studies initiated in 1996 and 2000 to address these issues.

MATERIALS AND METHODS

In 1996 and 2000, ponderosa pine sites were selected in the Modoc National Forest and near Poison Lake on the Lassen National Forest, respectively, in northeastern California. The first study (Modoc National Forest) objective was to determine effects of high impact and low impact thinning conducted in spring or fall seasons. Fifteen 2.5 ha plots, including unthinned controls, were located and marked for randomly assigned thinning treatments. Codominant tree age in the plots was approximately 100 years. Thinning treatments were conducted during spring and fall of 1995 and consisted of 1) low impact thinning involving rubber tired

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skidders on designated skid trails and chainsaw falling only, and 2) High impact thinning involving use of tracked shears and skidding at operator discretion. Each treatment was replicated 3 times in a random design.

Plots were visited every year post treatment until 2002. Thereafter, a final measurement to date was conducted on all plots during 2006. Data on symptoms and mortality were recorded for all years. Only dead or moribund trees with confirmed black stain root disease as determined by chopping into the root collar and observing characteristic black streaking were documented as mortality due to blackstain root disease.

In the second study conducted in the Lassen National Forest, we addressed effects of prescribed burning and subsoiling on black stain root disease development. The study design was a randomized complete block with four replications. Each treatment plot was 2.5 ha, with four treatments per block. Treatments were 1) underburning only, 2) subsoiling skid trails only, 3) underburning and subsoiling, and 4) untreated control. The entire study site was 80 ha and had been thinned one year prior to plot and treatment establishment. Prior to thinning, the average stand basal area was 263 ft² / acre (60 m² / ha) and the average QMD (quadratic mean diameter) was 7.9 inches (20 cm). Post thinning stand density was 121 ft² / acre (28 m² / hectare) with an average QMD of 14.8 inches (38 cm).

In addition to conducting 100 percent yearly surveys on each plot to record symptom development and mortality, experiments involving large woody root inoculations with *L. wagneri* var *ponderosum* were also conducted. These experiments were designed to provide information on the minimum amount of spores carried by insects necessary to start root infection. Spore suspensions containing 50, 500, 5,000, 50,000, and 3,050,000 spores were injected in artificial wounds created by coring to 2 cm depth in the xylem with a 4 mm diameter increment hammer. The spore suspensions were placed into roots of randomly selected trees in the burn only and control plots. Lesions, including sterile control wounds were measured after 9 weeks. During 2002, woody roots of trees with fire scorch damage and those without were inoculated with *L. wagneri*-infested 4 mm diameter cores. Roots were re-excavated after 9 weeks and lesions measured. Data on stem cambial sucrose synthase activity, a surrogate for determining stress via carbohydrate status of the trees, was also obtained during the 2002 and 2003 season as in Otrosina et al. (1999).

We also carried out insect trapping using Lindgren flight traps during the 2002-2003 seasons to determine treatment effects and potential relationships with subsequent disease occurrence. A cluster of four traps per plot was used and baited alpha pinene and ethanol. Traps were checked every 2-4 days during the flight season and trapped insects were counted and sorted by species.

RESULTS AND DISCUSSION

Modoc National Forest

Cumulative mortality for the 10 years since treatment initiation is presented in *Figure 1*. Excluding the control plots, the high impact thinning treatment conducted in the spring resulted in the most mortality. The low impact spring thinning did not have mortality due to blackstain root disease. This must be interpreted with caution because by chance, the assigned treatment on these plots happened to be on soils not favorable for black stain root disease development. There may be a correlation between certain soil series, vegetation types, and occurrence of the disease (Kliejunas – Otrosina 1998).

Cumulative Mortality Due To Black Stain Root Disease

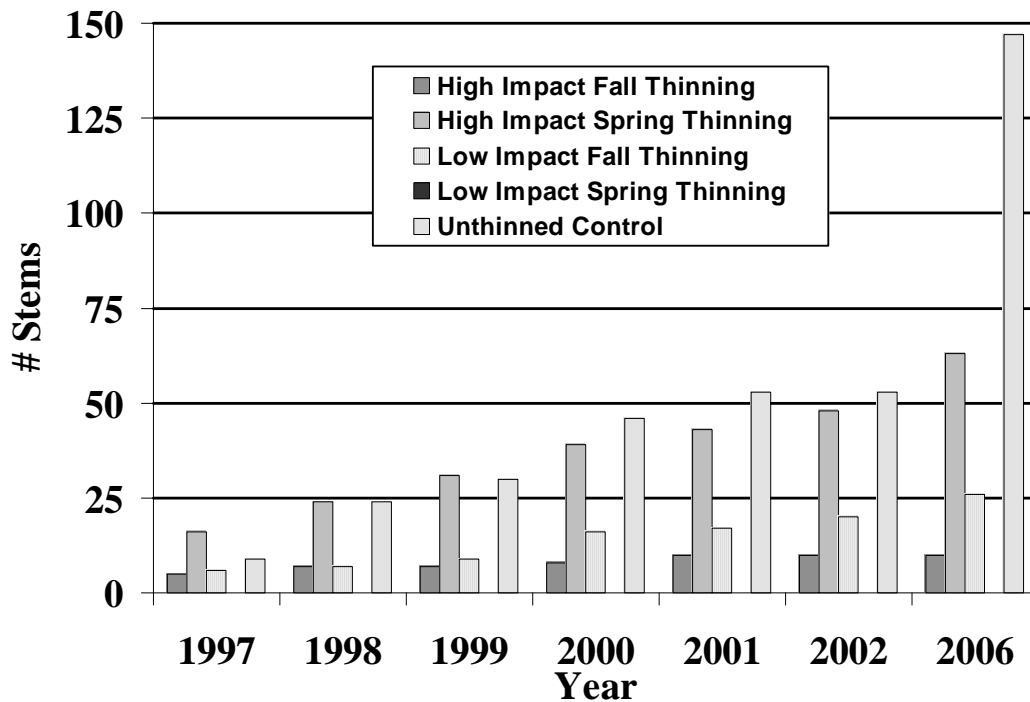


Figure 1. Mortality resulting from thinning treatments over 10 years following four thinning treatments. Note that low impact spring thinning plots never had blackstain root disease due to soil factors.

The control plots had dramatically more mortality than any of the thinning treatments. This is significant because it illustrates the benefit of lowering stand density and therefore stress in mitigating disease impact. Excessive stand density coupled with high mortality rates from black stain root disease can greatly increase risk of catastrophic wildfire in unthinned stands.

Lassen National Forest

While the study is long-term, intermediate results are interesting. Several root feeding insect species of interest, suspected to be potential vectors of *L. wagneri*, were caught during the two seasons. Among the more common species were *Hylastes macer*, *Hylurgops subcostulatus*, and *Hylurgops porosus*. Treatment differences in total insect trap catches are not obvious, although the underburn only plots tended to have slightly higher catches during the latter half of the flight season. In 2002, this trend appeared to be more marked, with greater catch numbers later in the season (Figure 2). Recently, DNA evidence indicated the insect species mentioned above, among others trapped on the study plots are carrying *L. wagneri*, presumably as spores (Schweigkofler et al., 2005). Such insect species have been suspected but heretofore have not been confirmed to be carrying *L. wagneri* in ponderosa pine stands. This confirms the long held notion that root feeding Scolytids serve as potential vectors of the fungus, critical for spread of the disease over longer distances.

Between 2001 and 2005, the burn only treatment had the highest mortality (Table 1). Scorching was evident on most of the mortality trees, which succumbed within two years following treatments. It has been approximately a century since fire last occurred in these stands. The subsoiling and burn treatment had considerably less mortality than the burn only

treatment. The subsoiled skid trails may have served to mitigate at least partially fire severity or intensity in these plots (*Table 1*). Consequently, caution should be exercised when reintroducing fire to stands that have not been burned for a considerable time.

Total Root-Feeding Bark Beetles Trapped in 2001

Total Root-Feeding Bark Beetles Trapped in 2002

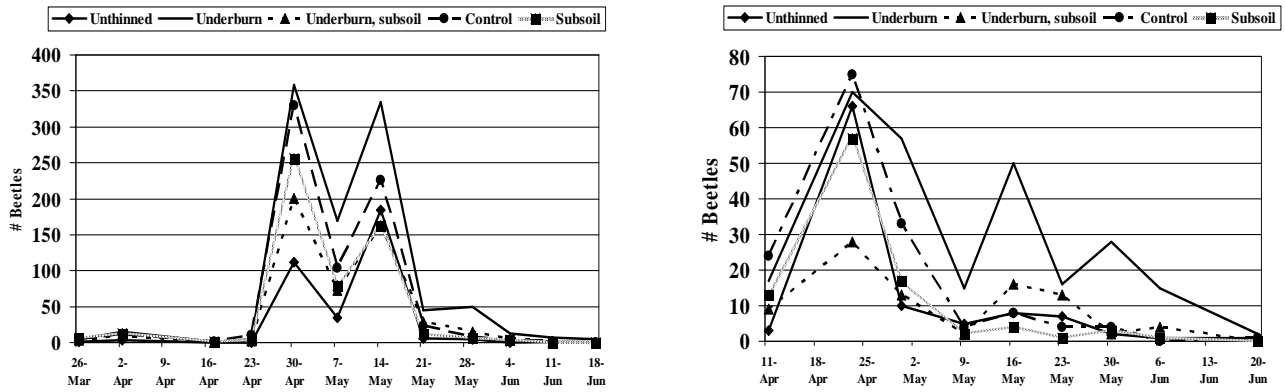


Figure 2. Summary of trap catches of root feeding Scolytids during the 2001 and 2002 trapping season. Note differences in scale between years.

Table 1. Poison Lake Mortality/Symptomatic Tree Data Summary

Treatment	Total # Dead Trees (2001-2005)	Symptomatic Trees	Confirmed Blackstain
Underburn	322 (40.00) ¹	18 (3.87)	11 (2.99)
Underburn, Subsoil	174 (40.00)	12 (2.16)	2 (1.00)
Subsoil	35 (10.24)	33 (8.54)	15 (6.18)
Control	37 (10.31)	25 (5.25)	8 (4.00)

¹ Standard deviation (n=4)

Findings in 2004 inoculation experiments using the three dosages of a local isolate of *L. wagneri* spores (Otrosina et al 2004) are presented in Figure 3.

2004 June Inoculations-Lesion Area

2004 August Inoculations-Lesion Area

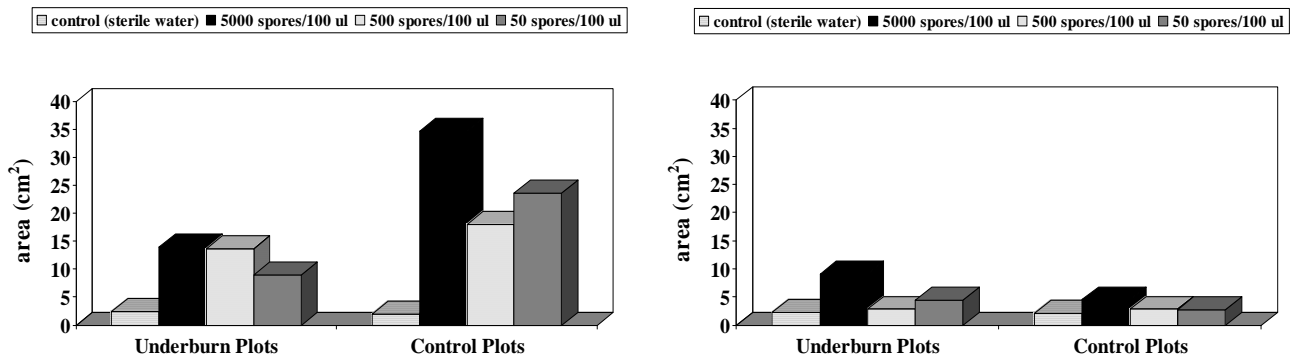


Figure 3. Lesion areas resulting from different *L. wagneri* spore dosages between underburned and control plots for June and August inoculations.

- 1) The June inoculations produced larger lesions in roots after 9 weeks than the August inoculation.
- 2) The lowest spore dose, 50 spores, produced lesions that were significantly larger than controls (June inoculations). This is noteworthy because it is consistent with the lower range of spore numbers found on potential insect vectors as determined by DNA analyses (Schweigkofler et al 2005).
- 3) Lesions in underburned plots trended smaller than control plots,
- 4) Lesions from August inoculations were significantly smaller than June inoculations
- 5) We recovered *L. wagneri* from lesions approximately one year (June 2005) after inoculation (June and August 2004) with the 5,000 spore inoculum dosages or mycelial inoculum.

Sucrose synthase activity, a measure of tree physiological status, shows a seasonal trend between the sampled months in 2003, and 2004. Peak activity is attained during July and August and drops rapidly during September. This is consistent with other data reported for ponderosa pine (Otrosina et al, 1996). These data seem to be negatively correlated with the lesion sizes in the June and August inoculations. The meaning of these relationships is unclear at this time but the physiological status of the tree and interactions with insect feeding, infection, and subsequent disease expression are important research topics that must be addressed.

In 2005, 100% surveys of each plot showed symptomatic trees, based upon crown characteristics, were distributed evenly among treatments, and few confirmed black stain root diseased trees were found. This is to be expected due to the longer time interval we anticipate from treatment initiation to infection, colonization, and symptom expression in the trees. Thus, further long-term monitoring of these study plots is necessary and planned.

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LITERATURE CITED

- HARRINGTON T.C. – COBB, F.W., Jr. (1988): *Leptographium* root diseases on conifers. American Phytopathological Society, St. Paul, MN. 149 p.
- KLIEJUNAS, J.T. – OTROSINA, W.J. (1998): Site factors associated with incidence of black-stain root disease in east-side pine. *Phytopathology* 88 (9):S48.
- OTROSINA, W.J. – FERRELL, G.T. (1995): Root diseases: Primary agents and secondary consequences of disturbance. pp 87-92. In: Eskew, L., Compiler. Forest Health Through Silviculture. Proc. 1995 National Silviculture Workshop, May 8-11, 1995; Mescalero, NM. USDA Forest Service Gen. Tech. Rept. RM-GTR-267, Fort Collins, CO. 246 p.
- OTROSINA, W.J. – SUNG, S-J. – WHITE. (1996): Effects of subsoiling on lateral roots, sucrose metabolizing enzymes, and soil ergosterol in two Jeffrey pine stands. *Tree Physiology* 16: 1009-1013.
- OTROSINA, W.J. – KLIEJUNAS, J.T. – SUNG, S.S. – SMITH, S. – AND CLUCK, D.R. (2004): Development of black-stain root disease on artificially inoculated ponderosa pine. *Phytopathology* 94: p. 79.
- SCHWEIGKOFER, W. – OTROSINA, W.J. – SMITH S.H. – CLUCK, D.R. – MAEDA, K. (2005): Detection and quantification of *Leptographium wagneri*, the cause of black-stain root disease, from bark beetles (*Coleoptera: Scolytidae*) in Northern California using regular and Real-time PCR. *Can. J. For. Research* 35: 1798-1808.