

Summer Survival of *Phytophthora ramorum* in Forest Soils¹

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Abstract

Recovery of *Phytophthora ramorum* from soils throughout Sudden Oak Death-affected regions of California illustrates that soil serves as an inoculum reservoir for the pathogen, but the potential for survival in soils throughout the summer is largely unknown. In this study we assess pathogen survival in infected leaf tissue in the upper soil profile in a redwood-tanoak forest ecosystem in Sonoma County, California.

Over 4,000 rhododendron leaf disks were inoculated with an isolate of *P. ramorum* that was recovered from soil at Jack London State Park. Inoculated leaf disks were incubated for one week in a moist chamber to allow for colonization of leaf tissue. Ten leaf disks were placed in each of 360 mesh sachets and transferred to the field in April 2004. The sachets were dispersed under 10 trees each of tanoak (*Lithocarpus densiflorus*), California bay laurel (*Umbellularia californica*), and coast redwood (*Sequoia sempervirens*), and at three vertical locations: i) leaf litter surface, ii) litter/soil interface, and iii) below the soil surface. Sachets were arranged vertically within partially submerged cylindrical barriers to prevent lateral movement of *P. ramorum* and were retrieved 1, 2, 8 and 24 weeks after introduction.

At each time point, sachets were retrieved from the field, and soil samples were taken from within the cylindrical barriers and from the bulk soil. In the laboratory, the wet weight of leaf disks was recorded as a measure of water retention; soil moisture content was also determined. *P. ramorum* was baited from soil samples, both inside the cylinders and from the bulk soil under each tree. Pathogen recovery was determined by submerging the leaf disks in selective medium and incubating at room temperature for 14 days. If a leaf disk was considered *P. ramorum*-negative, that disk was subsequently incubated in water for three weeks before being returned to selective medium. After determination of pathogen recovery, leaf disk samples retrieved after 24 weeks were cleared with 1 M KOH and chlamydospore populations were assessed within the tissue.

After one week in the field, the pathogen was recovered from 1 percent of the disks at the leaf litter surface. Recovery from the two subsurface treatments remained over 80 percent over the first 2 weeks. After 8 weeks 80 percent and 65 percent pathogen recovery were observed in the soil and at the

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litter/soil interface, respectively. Over 60 percent pathogen recovery was observed in leaf disks remaining in the soil for 24 weeks. After incubation of *P. ramorum*-negative disks in water, recovery at 8 weeks was enhanced by 3 percent and 10 percent in soil and interface disks, respectively. Significantly more chlamydospores were observed in leaf disks ranked *P. ramorum*-positive after incubation in water than those ranked *P. ramorum*-negative, and chlamydospore germination was occasionally observed within *P. ramorum*-positive leaf disks. Furthermore, leaf disks incubated in soil contained significantly more chlamydospores than those incubated at the surface or litter/soil interface.

Soil moisture content varied under the different tree species, with the highest moisture content persisting under redwoods, as well as tree species associated with organic soils. Soil moisture content was positively correlated with wet weight of leaf disks ($P \leq 0.01$), and leaf disk wet weight was significantly correlated with recovery ($P \leq 0.01$) over the first 8 weeks; however, no direct correlation between soil moisture content and pathogen recovery was observed. *P. ramorum* was baited from soil within 33 percent of the cylinders 1 week after introduction of inoculum, and from 16 percent of the cylinders at every subsequent sample time. Conversely, *P. ramorum* was rarely recovered from soil sampled outside the cylinders.

Heightened recovery of *P. ramorum* from soils after a period of oversummering suggests that infested soil may serve as a source of primary inoculum for fall disease development. The ability to bait *P. ramorum* from soils within cylinders demonstrates the transmission of the pathogen from infected leaf tissue to the bulk soil. Recovery of *P. ramorum* after incubation in water coupled with high populations of chlamydospores in water-soaked *P. ramorum*-positive tissue suggests the potential role of hydration in breaking chlamydospore dormancy. Further studies are needed to address the relationship between pathogen survival and soil moisture content and soil matric potential; however, the results of this study suggest that natural pedoturbation processes resulting in burial of inoculum may enhance pathogen survival.