

A Survey of *Phytophthora*, *Pythium*, and *Phytophthora* Species in Soil from Upland Prairie Restoration Sites in Western Oregon¹

Jennifer Parke,² Erika Mittermaier,² Neelam Redekar,²
Joyce Eberhart,² and Teresa Matteson³

Abstract

Native upland prairie and oak savanna habitats were once widespread in the Willamette Valley of western Oregon, but have been diminished by conversion to other land uses. These threatened habitats are considered essential for rare and endangered species such as the Fender's blue butterfly. Restoring native upland prairie habitats is a major goal of wildland restoration in Oregon.

The inadvertent spread of *Phytophthora* species from nurseries into native ecosystems can have long-term environmental and economic impacts, as has been seen with *Phytophthora ramorum*, *P. lateralis*, *P. cinnamomi*, *P. tentaculata*, and other species. The risk may be particularly great when nursery-grown plants infested with *Phytophthora* spp. are planted in restoration sites, introducing pathogens directly into native habitats (Garbelotto and others 2018). The objectives of this study were to survey the distribution of *Phytophthora*, *Pythium*, and *Phytophthora* spp. in upland prairie restoration sites in western Oregon and to determine if they are detected at greater frequency in planted vs. non-planted sites.

We collected soil samples (0-20 cm depth) from 55 upland prairie/oak savanna sites in the Willamette Valley in western Oregon between November 2016 and May 2017. Of these, 27 sites had been planted with seeds, bulbs, or live plants within the last 5 years, and 28 sites had not been planted. Soil was sieved (2-mm) and split into two homogenized subsamples. One set of subsamples was baited with pears. Pear lesions were plated to obtain pure culture isolates. DNA was extracted from cultures and species were identified based on Sanger sequencing of the ITS region. From the second set of subsamples, DNA was extracted directly from 10 g of soil. PCR was performed with these extracts using oomycete-specific ITS primers before sequencing on the Illumina MiSeq platform. Samples with low DNA (< 2ng/μl) were excluded. Species abundance was normalized across all samples and adjusted for the total amount of DNA recovered from the soil. A species was considered present in soil if its abundance was ≥1% of the sample DNA.

¹A version of the paper was presented at the Seventh Sudden Oak Death Science and Management Symposium, June 25-27, 2019, San Francisco, California.

² Dept. of Crop and Soil Science, Oregon State University, Corvallis, OR 97331.

³ Benton Soil and Water Conservation District, Corvallis, OR 97330.

Corresponding author: J. Parke, jennifer.parke@oregonstate.edu.

We detected 13 *Pythium* species, 5 *Phytophthora* species, and 1 *Phytopythium* species with the pear baiting and metabarcoding approaches (fig. 1). *Pythium* species were nearly ubiquitous, detected in 46 of the 55 sites (84%). *Pythium attrantheridium* was the most frequently detected species with the metabarcoding approach. It was not recovered from pear baits. Other *Pythium* species included the *glomeratum*-complex, *pectinolyticum*, *paroecadrum*, *macrosporum*, *ornacarpum*-complex, *volutum*, *pachycaule*-complex, *parvum*, *mamillatum*-complex, *terrestris*-complex, and *ultimum*. *Phytophthora* species were detected, but in only 7 of 55 sites (13%), and included the *P. cactorum* cluster, *P. megasperma* complex, *P. cambivora* complex, *P. fragariae* complex, and a *P. ramorum*-like species. A complex consists of closely related species that are indistinguishable based on the ITS1 region. A cluster consists of closely related species that are identical between the priming sites (Redekar and others 2019). Only two of the five *Phytophthora* species were recovered from pear baits. A quantitative real time PCR assay confirmed that the *P. ramorum*-like species has an ATP9-NAD9 region different than *P. ramorum*, despite an identical ITS1 region. There was no statistically significant relationship between the presence of *Phytophthora* ($p=0.389$) or *Pythium* ($p=0.503$) species and planting history based on contingency tables using a Pearson's test for independence (data not shown).

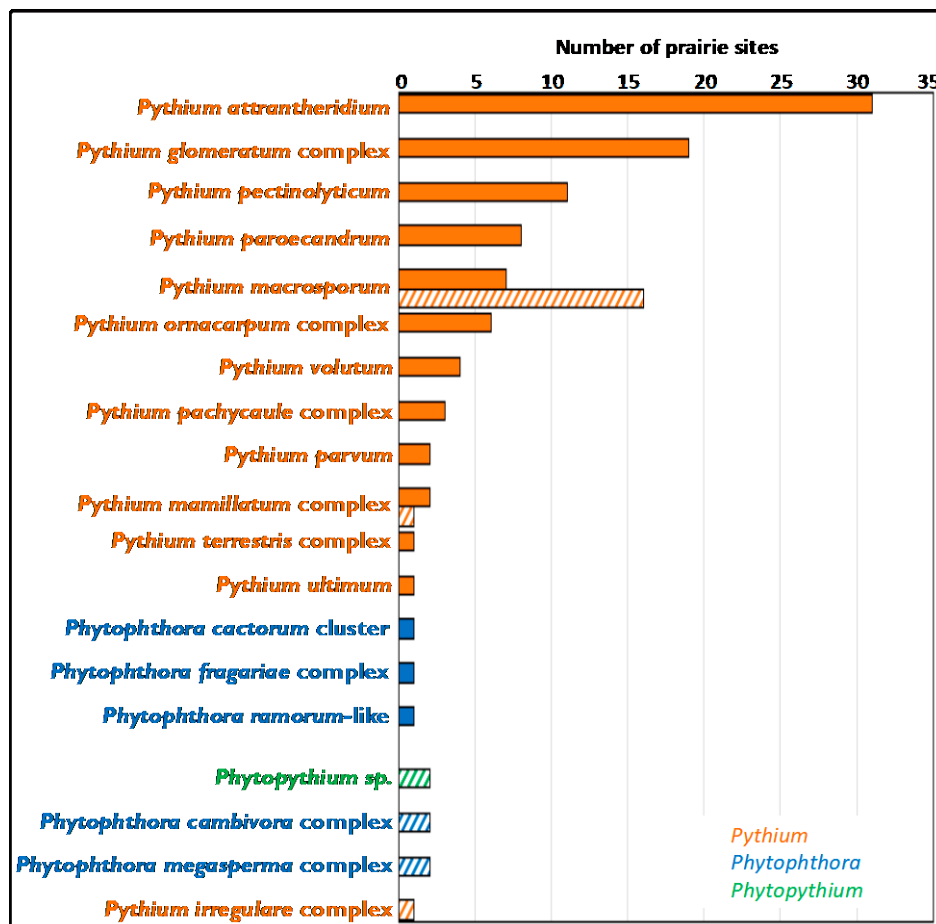


Figure 1—Frequency distribution of *Pythium*, *Phytophthora*, and *Phytopythium* species detected in soil from 55 upland prairie sites in western Oregon using pear baiting (striped bars) and Illumina MiSeq sequencing (solid bars).

Pythium appears to be nearly ubiquitous in upland prairie soils of western Oregon. The ecological role(s) of *Pythium* in this habitat is not known but *P. attrantheridium* has been shown to affect the distribution of native plants elsewhere (Allain-Boulé and others 2004, Packer and Clay 2000). In contrast, *Phytophthora* species were detected relatively infrequently in upland prairie soils but do include plant pathogenic species clusters or complexes (*P. cactorum* and *P. cambivora*) of potential concern to wildlands. Pear baiting resulted in detection of only 6 species. Illumina MiSeq appears to be a more sensitive method of detection, expanding the number of species or species complexes detected to 15.

Results of this study provide a snapshot of the current distribution of *Phytophthora* and *Pythium* species in restoration sites in western Oregon and can serve as a baseline for recognizing future introductions.

Literature Cited

- Allain-Boulé, N.; Tweddell, R.; Mazzola, M.; Bélanger, R.; Lévesque, A.C. 2004. *Pythium attrantheridium* sp. nov.: taxonomy and comparison with related species. Mycological Research. 108(7): 795–805.
- Garbelotto, M.; Frankel, S.J.; Scanu, B. 2018. Soil- and waterborne *Phytophthora* species linked to recent outbreaks in Northern California restoration sites. California Agriculture. 72: 208-216.
- Packer, A. and Clay, K. 2000. Soil pathogens and spatial patterns of seedling mortality in a temperate tree. Nature. 404: 278–281.
- Redekar, N.R.; Eberhart, J.E.; Parke, J.L. 2019. Diversity of *Phytophthora*, *Pythium*, and *Phytopythium* species in recycled irrigation water in a container nursery. Phytobiomes Journal. 3: 31-45.