## Phytophthora Diversity in Lake Mathews, the Irrigation Water Source for a Southern California Nursery<sup>1</sup>

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## Abstract

Lake Mathews is a 182,000 acre-feet capacity reservoir located in Riverside County, California. It is the western terminus of the Colorado River Aqueduct that serves as the main source of irrigation water for several horticultural nurseries in southern California. Some of these nurseries do not disinfest Lake Mathews water before using it for irrigation. This could increase disease pressure at these nurseries and facilitate survival, propagation and spread of important waterborne plant pathogens such as *Phytophthora* spp. on infested nursery stock.

We examined the diversity of *Phytophthora* species detected in this water over the span of 30 months to determine what risks it poses to the container nurseries that use this water source for irrigation. Metabarcoding approach: We periodically collected two 1-L water samples from Lake Mathews as supplied by the Western Municipal Water District to a large container nursery in southern California. One set of samples was filtered through 5µm Millipore nylon membranes to physically capture all *Phytophthora* species present in the water. Another set of samples was baited using rhododendron leaves to detect viable *Phytophthora* species in the water. In both cases, the ITS1 region was first amplified from the filter and leaf bait DNA using ITS6 and ITS7 primers (Cooke and others 2000), and then sequenced with high-throughput Illumina MiSeq 250PE sequencing for metabarcoding. The paired-end Illumina sequences were first cleaned, quality filtered and queried against a custom oomycete reference ITS database (Redekar and others 2019) using a nucleotide megablast search. Operational taxonomic units (OTUs) or taxa were identified based on percent sequence similarity to the reference sequence of known *Phytophthora* species.

In some cases, the Illumina MiSeq sequencing approach was incapable of differentiating between sequences of closely related species. Such closely related species were either classified as species complex or cluster, depending whether they share sequence identity across an entire ITS1 region, or across a shorter amplified ITS1 region, respectively.

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Twenty-two *Phytophthora* taxa were detected on filters; of these, 9 taxa colonized leaf baits (Figure 1). The ITS1 sequences originating from this study showed over 99% similarity to some of the important *Phytophthora* taxa including: *P. lateralis, P. citricola*-complex, *P. capsici*-cluster, *P. cryptogea*-complex, *P. citrophthora*-cluster, *P. tropicalis,* and *P. amaranthi*. We also detected ITS1 sequences that matched *P. ramorum* and *P. kernoviae*. Species-specific quantitative real time PCR assays (Bilodeau and others 2014) confirmed them to be similar to but not identical to *P. ramorum* and *P. kernoviae*, respectively. There were no seasonal trends in the occurrence of *Phytophthora* species detected in Lake Mathews water. Detection of most species was limited to a particular sampling time, where it was detected in greater abundance.

Lake Mathews water harbors plant pathogenic *Phytophthora* species that could pose disease risks at the nursery. It should be disinfested before use in irrigation. The metabarcoding approach allowed detection of *Phytophthora* species that were not recovered by baiting. Illumina MiSeq amplicon sequencing technology is very effective and sensitive for describing community composition, however its shorter sequences do not permit differentiation between closely-related species in some cases, resulting in unresolved species complexes or clusters.

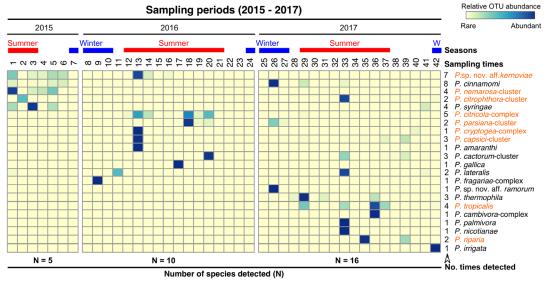


Figure 1 – Detection of Phytophthora in Lake Mathews water with metabarcoding. Species also detected on rhododendron leaf baits are indicated in orange.

## Literature Cited

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