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Disease on Nursery Stock as Affected by Environmental Factors and Seasonal Inoculum Levels of *Phytophthora ramorum* in Stream Water Used for Irrigation

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ABSTRACT

Tjosvold, S. A., Chambers, D. L., Koike, S. T., and Mori, S. R. 2008. Disease on nursery stock as affected by environmental factors and seasonal inoculum levels of *Phytophthora ramorum* in stream water used for irrigation. Plant Dis. 92:1566-1573.

A pear bait monitoring system was used to detect and quantify *Phytophthora ramorum* propagules in streams that flow through woodland areas with sudden oak death in Santa Cruz County, CA from 2001 to 2007. Stream propagules were detected most frequently or occurred in highest concentrations in winter and spring. The stream propagule concentration was characterized with statistical models using temperature and rainfall variables from 2004 to 2007. The highest concentrations of propagules occurred when stream sampling was preceded by about 2 months with low maximum daily temperatures and by 4 days with high rainfall. The occurrence of propagules in streams in the summer was mostly associated with infected leaves from the native host *Umbellaria californica* that prematurely abscised and fell into the water. When the stream water was used for irrigating rhododendron nursery stock from 2004 to 2007, disease occurred only three times in the two wettest springs (2005 and 2006) on plants sprinkler irrigated with stream water with relatively high concentrations of propagules. Disease incidence was described with a statistical model using the concentration of infective propagules as measured by pear baiting and consecutive hours of leaf wetness measured by electronic sensors at rhododendron height. The concentration of infective propagules was significantly reduced after water was pumped from the stream and applied through sprinklers.

Phytophthora ramorum is the causal agent of the disease known as sudden oak death (SOD). The pathogen causes trunk cankers and widespread mortality on tanoak (*Lithocarpus densiflorus*) and oak (*Quercus* spp.) (19), and leaf spots and blights on numerous other native hosts in California and Oregon woodlands (1,5). The pathogen was described as a new *Phytophthora* species in 2001, but it was observed as early as 1993 to cause leaf blights and mortality on rhododendron and viburnum in nurseries and public gardens in Germany and the Netherlands (27). With the recognition that this newly identified pathogen caused SOD, intensive nursery stock and public garden inspections ensued, and *P. ramorum* was found in several European countries. In December 2000, *P. ramorum* was first discovered infecting rhododendron nursery stock in California (8). By 2003, agricultural inspectors found the pathogen infecting nursery stock in California, Oregon, Washington, and British Columbia, Canada. In 2004, the disease became a national concern when a large wholesale nursery in California shipped camellia plants infected with *P. ramorum* to nurseries and other customers in 40 states. Presently, the Animal and Plant Health Inspection Service (APHIS) Plant Protection and Quarantine (PPQ) Agency lists 110 plant taxa as proven or associated hosts (1). There are various state, federal, and international quarantine restrictions placed on the movement of plants or plant parts of these listed hosts.

Once propagules are produced by *Phytophthora* spp. in natural environments, they can be moved from affected drainages through rain, runoff, and in streams as well as other means (7,18). Perennial and intermittent streams often run through areas of high incidence of SOD in California and Oregon woodlands, and *P. ramorum* has been detected in these streams (6,24). Stream water is sometimes used for irrigation by nurseries and landowners, and consequently propagules might be pumped from streams, moved through irrigation systems, and dispersed onto nursery or landscape hosts.

There are many important factors that should be understood to manage the risk involved with using contaminated irrigation water in a nursery or landscape. It is important to have an effective method to detect and quantify propagules in the stream and to understand the environmental conditions that are favorable to disease when stream propagules are present. We first monitored for the presence of *P. ramorum* propagules for over a year in several local streams flowing through areas where there was a high incidence of SOD to determine the general distribution of infested streams. Then for more than 4 years, we monitored propagule presence and concentration in one of these infested perennial streams. Concurrently, rhododendrons in an experimental layout that simulated nursery conditions were irrigated when needed with this stream water and noninfested domestic water. Each water source was applied with sprinkler and drip irrigation methods. The results from experiments will help determine the usefulness of stream baiting to detect and quantify propagules in streams and to assess the risk involved in irrigating with contaminated stream water, and provide information for the development of nursery management practices to control this disease.

MATERIALS AND METHODS

Pear baiting of stream samples to detect and quantify propagules. For the stream survey (2001 to 2002) and irrigation experiment (2004), the stream water sample was collected and baited with two green (unripe) D'Anjou pears. Pears were purchased as needed in small lots and stored under refrigeration until used. For the irrigation experiment (2005 to 2007), each of the three 8-liter stream water samples was baited with three pears. First, the pears were washed with liquid dishwashing detergent, rinsed, and allowed to dry. The pears were then suspended in each water sample so that half of each pear was submerged in the water by means of plastic bird netting secured to the bucket with elastic cords. The pears were incubated in the water samples for 24 h and then gently rotated to expose the entire pear to the water samples that potentially contained propagules. After another 24 h, the pears were removed from the water and placed on absorbent paper towels for an additional 48 to 72 h at a mean temperature of 20°C in open air and natural light. After some experience with subculturing, lesions caused by *Phytophthora* species could easily be separated from those lesions caused by *Pythium* species (as described in Results section). Tissue from the margin of

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