Susceptibility of Umbellularia californica to Phytophthora ramorum



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INTRODUCTION

Phytophthora ramorum is a newly described aerially disseminated phytopathogen that threatens the coastal oak forests in California (1). The coexistence of a number of different hosts in these forests allows the pathogen to become established. In particular, severe stem cankers on *Quercus agrifolia* (coast live oak) are strongly associated with heavily infested leaves of adjacent *Umbellularia californica* (bay laurel) (2). Often asymptomatic to low symptomatic bay trees are observed next to severely infested bays.

We designed a bioassay to screen for variation in susceptibility to *P. ramorum* among populations of bay trees. By understanding the conditions most conducive for infection of bays, we can determine which oak woodlands have the highest potential risk of infection by the pathogen.

METHODS

Bioassay development

Branches with 4 first expanded shade leaves were collected from various locations in California. Microcentrifuge tubes were attached to leaf tips (Fig. 1). To each tube, 300 μ L of zoospores (2x10⁴/mL) was added. Branches were incubated in moist biodomes for 2 weeks. Lesion area was measured.

Two optimization studies were conducted:

1. Exposure time: leaves were exposed to zoospores for 6, 12, 24, 36 and 48 h. Microtubes were carefully removed.

2. Temperature: after inoculation, leaves were incubated at 12, 17, 27 and 29 °C

Inoculation study

Branches from 6 trees (3 reps/ tree) were collected from 4 *P. ramorum* infested locations in California. The locations were Big Sur (Monterey), China Camp State Park (Marin), Swanton Pacific Ranch (Santa Cruz), and Austin Creek State Recreational Area (Sonoma). Branches were incubated as above, but at optimized exposure time (12 h) and temperature (17-22 °C).

RESULTS

Bioassay development

Exposure time: Leaves exposed to zoospores for 6 h had smaller lesions than those exposed for 12-48 h. Therefore, 12 h was used for the bioassay.

Temperature: Largest lesions formed at 17-22 °C. Towards the maximum and minimum temperature range, little or no differences were evident in lesion size among leaves collected from different trees.

Inoculation study

Lesions within a location varied significantly among trees (Fig. 3). Variation between locations was also significant (Fig. 2, 3). Overall, leaves from Swanton Pacific had significantly smaller lesions, while those from China Camp had significantly larger lesions.



Fig. 1. Method used to inoculate *Umbellularia californica* leaves on small branches with *Phytophthora ramorum* zoospores. Zoospores were added to microfuge tubes.



Fig. 2. Leaf tip lesions that developed 14 days after inoculation with *Phytophthora ramorum* zoospores on representative leaves of *Umbellularia californica* collected from 4 locations in California.

DISCUSSION

The bioassay is a reliable and practical screening system for assessing bay susceptibility. We found a range of susceptibility within and among infested locations in California. These and other locations will be assessed more intensively.

It is hypothesised that a number of factors contribute to *P. ramorum* epidemics including host resistance, genetic structure of host species in the forests, pathogen variation, and environmental conditions. These issues are currently being assessed in bay as well as *Q. agrifolia*.



Fig. 3. Average lesion area that developed 14 days after inoculation with *Phytophthora ramorum* zoospores on leaves of *Umbellularia* californica collected from 4 locations in California; n = 3.

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