Evaluation of Chemical and Biological Agents for Control of *Phytophthora* Species on Intact Plants or Detached Leaves of Rhododendron and Lilac¹

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Abstract

The recent incidence of Ramorum blight, caused by Phytophthora ramorum, on many nursery crops has focused attention on improving management strategies against Phytophthora diseases in nurseries. We evaluated several chemical agents that target Oomycete pathogens for their capacity to inhibit infection of rhododendron or lilac leaves by P. ramorum (both NA strain 2027 mating type A2 and European strain D12A mating type A1) compared to other species, including P. cactorum, P. citricola, P. nicotianae, and P. citrophthora. We inoculated needle-wounded leaves from plants previously treated with various chemicals that were (a) removed and inoculated, or (b) inoculated on intact plants maintained in high humidity. Inoculation of leaves on chemically-treated intact plants with P. ramorum or other *Phytophthora* species yielded similar results to those from inoculation of leaves detached from the same treated plants. Most of the chemicals tested had some efficacy on some species of *Phytophthora*, but Subdue Maxx (drench or foliar) had the greatest disease-suppressive activity against all species of Phytophthora except P. citrophthora, and was effective for at least 6 weeks after drench application. Some chemicals had varied efficacy depending on the species of Phytophthora. All chemicals were fungistatic, not fungicidal. Dipping leaves in the chemicals 24 hr prior to inoculation resulted in the same activity profile as applying chemicals to intact plants or detached leaves. Bacterial antagonists (Bacillus brevis or isolates of Paenibacillus polymyxa) significantly inhibited all *Phytophthora* species in *in vitro* challenges, but were ineffective when applied to leaves 24 hr prior to inoculation with P. ramorum or other species.

Key words: Phytophthora ramorum, sudden oak death, ramorum blight, chemical control, biological control

Introduction

Surveys in many areas of the world indicate that many species of *Phytophthora* can cause diseases in the nursery industry but are not being controlled by current management practices.

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That fact and the recent emergence of Ramorum blight in nurseries, caused by *P. ramorum*, suggest the need for improved control strategies, including application of chemical and biological agents. The purpose of these studies, therefore, was to evaluate a range of chemicals, specifically targeted to Oomycete pathogens, and some prospective bacterial antagonists that strongly inhibit *Phytophthora* spp. *in vitro*, for their ability to suppress infections by a selection of *Phytophthora* species, including *P. ramorum*, that can cause foliar blights on rhododendrons and lilacs.

Materials and methods

Inoculation and disease severity assessment. Leaf material used was from outdoor containergrown rhododendron 'Nova Zembla' and lilac 'Monge' plants. Three replicate leaves per fungal isolate were inoculated with mycelial inoculum plugs taken from agar cultures of the different species of *Phytophthora* and placed directly on needle wounds. Inoculated detached leaves were placed in zip-lock polyethylene bags and hand-misted with water as the bags were closed several times during incubation. Leaves on intact plants were similarly inoculated and the entire plant enclosed in a polyethylene bag. All inoculated plants or leaves were maintained in an incubator at 20 °C for 8 days, and then leaves were photographed to obtain digital images from which percentage of leaf area with lesions was assessed quantitatively using the lesion assay software ASSESS (Lakhdar Lamari, APS, St. Paul, Minn.).

Chemical and biological agents. Eleven chemicals plus a control were tested for efficacy in controlling six species of *Phytophthora* inoculated onto rhododendron or lilac leaves. Chemicals used were: BAS 500, Agriphos, Aliette, Biophos, Fenamidone, Fosphite, Heritage, Ranman, Stature, Subdue Maxx, and Truban, all at label rates. Chemical agents were applied either by: (a) spraying foliage or drenching roots of container plants, or (b) by dipping detached leaves removed from container-grown plants. Bacteria were isolated from organic materials and tested *in vitro* against numerous *Phytophthora* species; all showed strong antagonism. Bacterial isolates were identified by FAME analysis (Texas A & M University) as *Bacillus brevis* and *Paenabacillus polymyxa*. Leaves to be inoculated were dipped in the bacterial suspensions and held for 24 hr prior to inoculation with the *Phytophthora* pathogens. Pathogenic *Phytophthora cactorum*, *P. citricola*, *P. citrophthora*, *P. nicotianae* (= *P. parasitica*), and two isolates of *P. ramorum* were grown on dilute V8 juice agar medium for 14 days before plugs were removed as inoculum.

Results and discussion

Inoculation of leaves on chemically-treated intact plants with P. ramorum or other *Phytophthora* species yielded similar results to those from inoculation of leaves detached from the same treated plants. Most of the chemicals tested had some efficacy on some species of Phytophthora, but Subdue Maxx (drench or foliar) had the greatest disease-suppressive activity against all species of *Phytophthora* except *P. citrophthora*, and was effective for at least 6 weeks after application. Some chemicals had varied efficacy depending on the species or isolate of *Phytophthora* and the host used. Dipping leaves in the chemicals 24 hr prior to inoculation resulted in the same activity profile as applying chemicals to intact plants or detached leaves, and in some cases efficacy was increased. Bacterial antagonists (Bacillus brevis or isolates of Paenibacillus polymyxa) significantly inhibited all Phytophthora species in *in vitro* challenges, but were ineffective in inoculation assays of leaves dipped in a cell suspension of each antagonist 24 hr prior to inoculation with P. ramorum or other species. These tests indicate that inoculating detached leaves was comparable to inoculating intact plants to evaluate chemical and biological agents against *Phytophthora* species, and that several systemic or translaminar chemicals were effective in suppressing infections but without eradicating the pathogens.



Figure 1 – Disease severity on chemically-treated rhododendron or lilac leaves inoculated with isolates of *Phytophthora ramorum* (isolates 2027 or D12A) or *P. citricola*. Bars on columns represent 95 percent confidence intervals.