CONTROL OF *PHYTOPHTHORA LATERALIS* ROOT ROT OF LAWSON CYPRESS WITH ENTEROBACTER AEROGENES

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Abstract. The biological agent strain B8 of *Enterobacter aerogenes,* applied in June of 1992, 1993, 1994, and 1995 as a soil drench at 6.7 x 10^8 colony forming units per tree, was evaluated for control of *Phytophthora lateralis* root rot of Lawson cypress (*Chamaecyparis lawsoniana*) trees. The test was conducted under field conditions in the city of Vancouver, British Columbia, on naturally infected Lawson cypress trees. Based on four years of observations, strain B8 of *E. aerogenes* controlled the disease and increased the growth of trees. This study showed the potential of strain B8 of *E. aerogenes* for biological control of root rot of Lawson cypress trees.

Introduction

The most important disease of Lawson cypress (*Chamaecyparis lawsoniana*, A. Murr., Parl.) is the root rot (1) caused by soil-borne fungus *Phytophthora lateralis* (Tucker & Milbrath). Trees of all ages are susceptible. The fungus attacks and destroys the roots and spreads into the base of the stem or trunk, giving the killed inner bark a cinnamon colour, with a sharp line of demarcation between necrotic and healthy bark. The role of Lawson cypress as an important forest and ornamental tree in the Pacific Northwest is threatened by the increasing impact of *P. lateralis* root rot.

Chemical treatment is not an option as there are no fungicides registered for this use in Canada. Lawson cypress seems to be incompatible with resistant rootstocks (2). Methods recommended for control of Lawson cypress root rot include growing disease-free stock in virgin soil, avoiding monoculturing of this tree in windbreaks or hedges, removing and destroying infected plants along with the entire root system, and ensuring adequate drainage.

Biological control of a related pathogen *Phytophthora cactorum (Lebert and Cohn) J. Schroet.* which causes crown and root rot of apple trees in the Okanagan and Similkameen Valley of British Columbia (B.C.) has been achieved with a B8 strain of *Enterobacter aerogenes* (5, 6). This strain was isolated from the local soil in Kelowna, located in the Okanagan Valley, B. C., Canada. The present study intended to evaluate the efficacy of the same strain of *E. aerogenes* as drench applications for control of Lawson cypress root rot and to study the effects of this treatment on tree growth.

Materials and Methods

Biological agent evaluated. The material evaluated was strain B8 of *E. aerogenes* developed at the Agriculture and Agri-Food Canada Research Centre, Summerland, B.C.

Field tests. The test was conducted in the city of Vancouver, B.C. on approximately 80- year-old Lawson cypress trees some of which showed typical symptoms of root rot caused by natural infection with *P. lateralis.* Twenty eight trees selected for the study were growing in the centre lawned median along King Edward Avenue. These trees were assigned a disease rating in February, 1992 (0=healthy—no infection at all; 1= initial—less than one-fourth of the roots at the crown region infected by *P. lateralis*; 2= intermediate—about one-fourth to one-half of the roots infected; 3=terminal—more than half the roots infected; 4=dead—all roots are infected, and the tree is dead). These ratings are similar to the ratings used for rating apple trees infected with *P. cactorum* (6). Bark and root samples were collected to determine infection by *P. lateralis.* The presence or absence of *P. lateralis* in bark and roots of infected trees was confirmed before the application of the biological treatment by culturing 10 subsamples / tree following the method by Matheron (3). Segments of decayed roots and bark 3 to 5 mm long were rinsed in tap water, dipped in 70% ethanol, dried on a paper towel, and pressed into a selective medium containing corn meal agar amended with 5 mg of pimaricin,

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Table 1. Effect of strain B8 of *Enterobacter aerogenes* on disease rating (0 = healthy; 4 = dead), and increase in trunk cross-sectional area (TCA, cm²) when applied as a soil drench from 1992-95 under park conditions.

	DISEASE RATINGS (0 - 4)			INCREASE IN TCA (sq. cm)
TREATMENT	1992	1995	MEAN[y]	
E. aerogenes (B8)	1.17 a[z]	0.93 a	1.06 a	621.46 a
Control	1.14 a	1.67 b	1.41 b	406.16 b
S.E.	0.17	0.17	0.15	46.75

[z]Means in the same column followed by the same letter are not significantly different (P = 0.05) according to the new Duncan's multiple range test.
[y]Means of both rating dates.

300 mg vancomycin hydrochloride, and 25 mg of pentachloronitrobenzene per litre. The plates were kept in darkness at 21°C and examined daily for 5-7 days for growth of *P. lateralis*. Colonies of *P. lateralis* were examined and confirmed microscopically.

Two treatments were replicated 14 times in a randomized block design. The treatments were 1) *E. aerogenes* applied as a drench in mid-June of 1992, 1993, 1994, and 1995, and 2) an untreated control. The same trees received approximately the same dosage annually. Control trees each received 20 litres of tap water. Suspensions of *E. aerogenes* (B8) were prepared (5) as follows. Bacterial growth from each culture plate after 48 hours was scraped into 100 ml of sterile water. Eight-ml bacterial suspensions, which were diluted in 20 litres of tap water corresponded to approximately 6.7 x 10^9 colony forming units (CFU) and applied to each tree on each treatment date. The drench applications were made by spraying the trunk to a height of about 60 cm above ground level and the ground within a 1 m radius around the trunk.

The diameter of each tree (1 m above ground) was measured in the third week of February, 1992, and again in late July of 1995. Increases in trunk diameter were calculated by subtracting the 1992 from the 1995 measurement. The values for increases in trunk diameters were converted to trunk cross-sectional areas (TCA) for statistical analysis. A disease rating (0= healthy, 1=initial, 2=intermediate, 3=terminal, 4=dead) was also assigned to each tree in July of 1995. The incidence of root or bark rot in 1995 was determined by culturing bark and root samples as described above for 1992. All data were analyzed by General Linear Model (GLM) procedures (4).

Results

Phytophthora lateralis was isolated from bark and root samples of all trees showing typical symptoms of crown and root rot. However, this fungus was not found in non-symptomatic trees.

There was no significant difference for disease ratings between the trees treated with biological agent *E. aerogenes* and untreated control trees prior to application of the treatment in 1992 (Table 1). However, a significant difference was observed between treated trees and untreated control trees in 1995. Similarly, a significant difference was observed between treated and untreated trees when the data were pooled for 1992 and 1995.

For both calculations, the trees treated with the biological agent *E. aerogenes* had significantly lower disease ratings than the untreated control trees.

No significant difference was observed for trunk cross-sectional area (TCA) between treated and control trees in 1992. However, a significant difference was observed for increases in TCA between treated and nontreated trees. The trees treated with *E. aerogenes* had significantly larger TCA than the untreated control trees.

Discussion

In the absence of a proven, economical method of preventing Lawson cypress root rot, it is probable that early diagnosis and timely curative treatment will remain the most practical and economic ways of dealing with this disease. Our results indicate that the Lawson cypress trees treated with drench applications of strain B8 of *E. aerogenes* for four years had significantly lower disease ratings and had significantly higher TCA than water-treated controls. These observations are similar to the findings of Utkhede (5) and Utkhede and Smith (6), where the application of strain B8 of *E. aerogenes* significantly reduced disease symptoms caused by *P. cactorum* crown and root rot of apple trees in southern British Columbia.

Lawson cypress trees in B. C. parks generally experience significant annual loss from *P. lateralis* root rot. The costs of replacing established Lawson cypress trees infected with root rot are increasing and may eventually become prohibitive. Moreover, new trees planted on sites formerly occupied by infected Lawson cypress trees are likely to suffer from the same pathogen unless the soil is treated. Biological treatment for root rot of Lawson cypress trees with strain B8 of *E. aerogenes* could become an alternative, environment-friendly treatment.

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