# Evaluation of Three Treatments for Eradication of *Phytophthora cinnamomi* from Deep, Leached Sands in Southwest Australia

TOM C. J. HILL, JOANNA T. TIPPETT, and BRYAN L. SHEARER, Research Centre, Department of Conservation and Land Management, Hayman Rd., Como, Western Australia, 6152

#### **ABSTRACT**

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Three treatments were evaluated against localized infections caused by Phytophthora cinnamomi in tracts of species-rich Banksia woodlands and scrub-heaths in deep, leached sands in southwest Australia. The treatments were (1) application of herbicide to kill all vegetation, (2) herbicide plus 5 or 15 g (a.i.) m<sup>-2</sup> metalaxyl, and (3) herbicide plus fumigation with 27 or 54 L m<sup>-2</sup> of 1% formaldehyde. The effectiveness of the treatments was assessed in 225-m<sup>2</sup> plots in two Jandakot sand sites using buried pine branch plugs inoculated with P. cinnamomi at 0.3 m and 1.3 m depth, and from naturally infected soil cores taken to 0.4 m depth. After 20 mo, P. cinnamomi was recovered from 77 and 87% of plugs buried at 0.3 and 1.3 m depth, respectively, in the herbicide plots, and from 4.6% of soil samples (21% in controls). With metalaxyl treatment, plug recovery was nil at 0.3 m and 3% at 1.3 m after 9 mo, and in soil samples taken after 10 and 20 mo, recovery was also nil. Formaldehyde efficacy varied with site; on one site, plug recovery was 4 and 2% at 0.3 and 1.3 m, respectively. From isolations from collar tissue of Banksia attenuata and adjacent soil samples, the fungus was retrieved from 100% of dying trees, 96% of recently dead trees and 56% of trees dead for more than 1 or 2 yr. In recently dead trees, fungal recovery from tap roots at 1.0 and 2.0 m depth was 87 and 48%, respectively. The longevity and occurrence of P. cinnamomi at deep depths precludes any attempt at rapid chemical eradication, although metalaxyl could be used to contain highly contagious sites. Confinement or eradication of the pathogen may be achieved by keeping sites completely bare for many years.

Widespread and accelerating destruction of heaths, woodlands, and forests by the introduced fungal pathogen Phytophthora cinnamomi Rands (A2 compatibility type) in southwest Australia (24) necessitates the development of methods to control new outbreaks of the disease in remaining tracts of healthy vegetation (19). Even though the pathogen is destroying many plant communities over a vast area (from Eneabba, 250 km north of Perth, to Cape Arid, 750 km to the southeast of Perth), large tracts of disease-free vegetation still remain. In particular, the Fitzgerald River National Park and the Lesueur National Park, both of which lie within nodes of extraordinary species richness and endemism, contain many highly susceptible communities that have largely escaped infection. Quarantine of these

Present address of first author: Department Of Environmental Sciences, University Of East London, Romford RD., London, E15 4LZ; present address of second author: PO Box 54, Walpole, Western Australia, 6398.

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areas will not ensure their permanent protection since the pathogen will eventually arrive. An active form of control, through eradication or confinement of small new infections, needs to be developed.

In Western Australia, the most endangered communities are woodlands and scrub-heaths that are dominated by three susceptible families (Proteaceae, Myrtaceae, and Epacridaceae). These include Banksia attenuata R. Br. and B. menziesii R. Br. low woodland, B. attenuata scrub-heath, B. coccinea R. Br. scrub-heath, B. baxteri R. Br. and Lambertia inermis R. Br. scrub-heath, and B. speciosa R. Br. scrub-heath. The soil profile common to all communities is deep, highly leached, and composed of medium-grained, quartz sand. This soil type occurs principally within a narrow coastal fringe of ancient, windformed dunes, and to a lesser extent farther inland where it was formed by erosional deposition on lateritic sandplains (2).

Any attempt to eradicate localized *P. cinnamomi* infections is aided by several attributes of disease epidemiology. First, the infected area can be defined readily by symptoms in the vegetation. A diseased site shows near total death of the

dominant tree or shrub species and heavy losses to the understorey, enabling the boundary between healthy and infected vegetation to be delineated sharply by a front of dying plants (7). Second, infections tend to expand slowly and predictably at  $1.0-1.3 \text{ m yr}^{-1}$  (7,17). This rate of pathogen spread suggests that the fungus is advancing by growing along the root systems of susceptible species, with cross infection occurring between plants in the interwoven network of lateral roots in the upper meter of the soil profile (7). The potential dependency of the fungus upon this network could be exploited to control localized infections. If the network is disrupted by killing all the vegetation on and around the site, further spread of the pathogen could be arrested. Further, if the site is kept completely bare, the fungal population may decline steadily. Fungicides could be used to lower the inoculum potential, thereby minimizing the potential spread of the fungus in contaminated

The objective of this research was to evaluate the effects of three treatments on the rate of reduction of *P. cinnamomi* in deep, leached sand beneath diseased *Banksia* woodland. All of the treatments entailed elimination of all vegetation on the site, along with application of a fumigant (formaldehyde) or a fungicide (metalaxyl) to eliminate the fungus to an appreciable depth (1.3 m) in the soil. In addition, the persistence of *P. cinnamomi* was measured in the tissue and surrounding soil of naturally infected *Banksia* stumps.

## MATERIALS AND METHODS

The study was sited near Gnangara, 30 km north of Perth (31°45'S., 115°52'E.), which lies within the Bassendean Dune System, a subunit of the Swan Coastal Plain. The Bassendean Dune System consists of low hills of highly leached, siliceous sands with intervening seasonal swamps where the near-level aquifer intersects the land-scape (13).

Persistence of P. cinnamomi in B. attenuata. Phytophthora cinnamomi survival was measured in the root systems and rhizospheres of dying and dead

B. attenuata in three diseased woodland sites. Trees were assigned to one of three categories according to their approximate age since death: (1) dying; (2) recently dead with leaves still attached (ranging from a complete crown to a sparse covering; progression from dying to leafless took between 1 and 2 yr, with trees beginning to lose leaves after 3 mo); and (3) long dead trees with no leaves remaining (dead for more than 1-2 yr). All dying trees lay along disease fronts and were chosen at random following yellowing of the crown, which usually occurred within a few weeks of the fungus girdling the collar region.

At each site, 7 dying, 15 recently dead, and 7-10 long dead trees were sampled. Three 1-L soil samples were taken from the top 10 cm of the profile at equidistant points around, and adjacent to, the collar, and were bulked. Using an axe, bulked samples of bark and then sapwood were taken from the same three points. The stump was excavated and soil and bark and wood samples were taken in the same manner at 30 cm depth. All dying and 7 recently dead trees on each site were excavated further, and samples were taken from 1.0 and 2.0 m depths. At each depth, soil was sampled, as described above, adjacent to the tap root and then a 10-cm length of tap root was removed with a saw. The average widths of the collar at the surface and at 30 cm depth, and of the tap root at 1.0 and 2.0 m depth, were 14.2, 9.5, 3.7, and 2.5 cm, respectively. Implements were sterilized with 70% alcohol between samples.

The soil samples were stored in sealed plastic bags, at room temperature (25 C), for a maximum of 2 wk before a 100ml subsample was tested for presence of P. cinnamomi using the Eucalyptus sieberi L. Johnson cotyledon baiting technique (12). Bark and wood samples were kept in separate plastic bags at 10 C for a maximum of 3 days before each sample was cut into small pieces and a subsample of each tissue type plated onto one 15-cm plate of selective medium (22). Plates were incubated at 26 C and examined with a microscope after 2 and 7 days for presence of  $\bar{P}$ . cinnamomi hyphae (12). Tools used for cutting and plating were sterilized with 70% alcohol between samples.

Evaluation of glyphosate, metalaxyl, and formaldehyde. The experiment was replicated at two sites, one of which was also used for the fungal persistence experiment. At each site, eight  $15 \times 15$  m plots were delineated in diseased vegetation along the dieback front. Two plots were randomly selected for each treatment, and the two remaining plots were controls. Plots were prepared by felling the *Banksias* and removing any stumps and vegetation from the plot surface (with a backhoe), and raking to remove remaining debris. A 1.5 m-deep trench

was dug around the perimeter of each plot, which was lined with a continuous sheet of black plastic and backfilled. Plots to be treated with formaldehyde were also bisected by a 0.5-m deep plastic-lined trench. A soil wetting agent (Once-wet, Langley Chemicals, Perth, Western Australia) was applied to all treatment plots at four times the recommended rate (0.02 L m<sup>-2</sup>) to reduce the strong nonwetting properties of the top 50 cm of the profile.

The following three treatments were applied in early spring (August and September 1989): (1) Herbicide. All remaining vegetation was killed by spraying with glyphosate at a rate of 7.4 kg (a.i.) ha<sup>-1</sup>. Glyphosate was reapplied after 4 and 12 mo to keep the plots completely bare; (2) Herbicide plus metalaxyl. Plots were treated with herbicide as outlined above. One-half of each plot was then treated with 5 g (a.i.) m<sup>-2</sup> of metalaxyl (Ridomil 5G granules, Ciba-Geigy Ltd., Melbourne, Victoria) and the other half with 15 g (a.i.) m<sup>-2</sup>; (3) Herbicide plus formaldehyde. Plots were treated with herbicide as outlined above. One-half of each plot was then sprayed with 27 L m<sup>-2</sup> of 1% formaldehyde, and the other half with 54 L m<sup>-2</sup>. Fumigated plots were covered with black plastic for 2 wk following application.

Monitoring of pathogen levels. One month prior to the application of treatments, P. cinnamomi was introduced systematically into the plots by burying colonized pine plugs at two depths. Pine plugs were made from debarked young Pinus radiata Ait. branches, 0.8-1.5 cm diameter, cut into 2.0-3.0 cm lengths (average fresh weight of 3.1 g) and with a small hole drilled through each. After autoclaving, plugs were inoculated with P. cinnamomi A2 (IMI 264384) grown on CMA and incubated for 1 mo at 26 C. By 3 wk, thick, aerial mycelium covered the plugs; prior to burial, two pieces from each flask were plated onto selective medium to confirm presence of the pathogen. Each plug was threaded onto a separate length of 70-kg breakingstrain nylon line and securely attached.

Plugs were buried in a randomly positioned systematic grid pattern of 32 points, composed of four lines of eight burial points, in each plot. Separation between grid lines was 4 m and between points on a line, 1.75 m. At each point, 3-4 plugs were buried, each spaced 0.25 m apart along an axis perpendicular to the grid line. All plugs at each point were buried at either 0.3 or 1.3 m depth. Deep burial points alternated with shallow along both axes of the grid. Each plug was pushed into the base of a shaft created by removing a 2.5-cm-diameter core of soil with a Veihmeyer tube. The shaft was then refilled with sand from the same soil horizon, leaving a length of the attached nylon line buried near the surface for later retrieval with a winch. Control, herbicide, and formaldehyde plots each contained 48 plugs at 0.3 m and 48 plugs at 1.3 m, while metalaxyl plots contained 64 plugs at each depth. When the plots were sampled, at least one plug was recovered from every point in each grid.

Survival of P. cinnamomi in buried pine plugs was assessed at 2-20 mo after application of the treatments. All plugs were retrieved from formaldehyde plots 2 mo after fumigation, and no further assessments were made since any residual activity of the chemical was assumed to be minimal due to its volatility and due to heavy spring rain. Plots treated with metalaxyl, which was assumed to have moderate residual activity, were assessed after 2 and 9 mo; one-half of the plugs were recovered on each occasion. In herbicide and control plots, one-third of the plugs were retrieved at each assessment (after 2, 9, and 20 mo). Recovered plugs were washed in single-distilled water and stored overnight at 10 C in moist, sealed plastic bags, with each plug from fungicide and fumigant plots stored separately. They were then surface sterilized in 70% alcohol for 30 sec, split in half, and the exposed faces plated onto P5ARPH selective medium (8). A separate plate was used for each plug originating from formaldehyde plots. After 10 days, all negative plugs were removed and re-tested for P. cinnamomi presence by re-splitting each half and plating the two newly exposed surfaces.

Soil population levels of P. cinnamomi were also measured in control, herbicide-, and metalaxyl-treated plots at intervals during the experiment. Formaldehydetreated plots were not tested due to formaldehyde's relatively low efficacy against inoculated plugs. Sixteen 1.25-L soil samples were taken from each sampled plot, and were composed of three bulked cores taken between 0.1 and 0.4 m depth. Soil samples recovered from 1.3 m depth tended to become contaminated with soil from overlying layers, and so were not taken. Fungal presence was detected using the cotyledon baiting technique as described above.

**Data analysis.** Prior to analysis, the proportional data produced by the presence/absence rating of each sample was normalized (arcsin of the square root). The influence of site, tree age, sample type, and depth upon *P. cinnamomi* persistence in *B. attenuata* was tested with multifactor analysis of variance using Tukey's honestly significant difference test for multiple range tests (21).

In the eradication experiment, *P. cinnamomi* survival was also expressed as the proportion of soil or plug samples that tested positive for fungal presence. Replicate plot results from each site were combined. The significance of the difference between proportions was deter-

Table 1. Characteristics of woodland sites used for measuring persistence of Phytophthora cinnamomi in Banksia attenuata stumps

Site	Topographic position	Soil profileª	Height above water table (m)	Vegetation type <sup>b</sup>
Gnangara Fire Tower	Upper slope	Jandakot sand	20–40	G (Leucopogon conostephioides DC., Scholtzia involucrata (Endl.) Druce, Eremaea pauciflora (Endl.) Druce, Melaleuca scabra R. Br., Astroloma xerophyllum (DC.) Sonder)
St. Patricks Road (Site 1 in eradication experiment)	Mid to lower slope	Jandakot sand	6–13	G (as above)
Lindley Road	Drained flat	Gavin sand	2–3	H (L. conostephioides, S. involucrata, Xanthorrhoea preisii Endl., Dasypogon bromeliaefolius DC.)

<sup>&</sup>lt;sup>a</sup> As defined by McArthur and Bettenay (13).

<sup>&</sup>lt;sup>b</sup> Banksia attenuata and Banksia menziesii low woodland as defined by Havel (6), with indicator understorey species in parentheses.

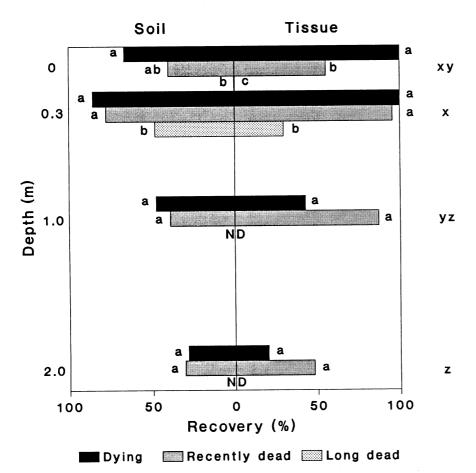


Fig. 1. Recovery of *Phytophthora cinnamomi* from tissue and adjacent soil samples taken from collar and tap root of dying, recently dead (leaves still attached), or long dead (no leaves remaining) *Banksia attenuata*. Combined data from three sites is shown (n = 21 trees for each horizontal bar, except for 0 m and 0.3 m samples from recently dead trees for which n = 45). Dissimilar letters denote statistical differences between groups: a, b, c for differences between time since death, determined separately for tissue and soil at each depth category; and x, y, z for the influence of sample depth on recovery rate, determined for combined tissue and soil samples of dying and recently dead trees (P = 0.05). ND indicates no data.

mined using the two-tailed Fisher's exact test (14).

## RESULTS AND DISCUSSION

Persistence of *P. cinnamomi* in *B. attenuata*. Topographic details of the sites used for tree excavation are given in Table 1. Adjacent, disease-free vegetation at each site was covered by *Banksia* low woodland (6–8 m high, and with a crown cover of 10–30%) dominated by *B. attenuata* and *B. menziesii*,

with B. ilicifolia R. Br., Eucalyptus todtiana F. Muell., and Nuytsia floribunda Labill. as minor components. A dense understorey of sclerophyll shrubs grew beneath the canopy.

The pattern of P. cinnamomi invasion of B. attenuata, and its subsequent persistence in tissue and surrounding soil, was similar in the three sites; analysis of variance revealed no effect of site upon overall fungal recovery rate (P = 0.89). Results from the three sites were there-

fore pooled (Fig. 1). Data from bark and wood samples were also combined since the fungus was recovered equally well from each tissue type. Recovery rate of P. cinnamomi was slightly higher in tissue than in soil samples, and although the difference was not significant when tested with analysis of variance, a paired sample Student's t test revealed a significant bias (P=0.01) toward tissue samples. Tissue and soil recovery data is therefore presented separately.

Large lesions that extended above ground level by an average of 0.5 m, although by as much as 1.5 m, were visible in the inner bark of all 21 dying trees. Phytophthora cinnamomi was recovered from all samples taken from the tissue at the soil surface and at a depth of 30 cm from these trees. When results from tissue and soil samples at 0 and 30 cm were combined and expressed simply as collar region recoveries, the fungus was also retrieved from 96% (n = 45) of recently dead trees (leaves still attached) and 56% (n = 27) of long dead trees (no leaves remaining). Although sample age had a highly significant influence upon recovery rate (P = 0.001), only the oldest category (dead for more than 1 or 2 yr) had a significantly lower residual P. cinnamomi population. In some of the long dead trees, wood-rotting fungi had noticeably decayed and bleached the bark and outer wood.

Fungal recovery from tissue and soil declined with increasing sample depth (P = 0.01), reflecting the mode of infection of the root system. Girdling of the collar region of 17 out of 21 dying B. attenuata excavated to 2 m depth had resulted from the invasion of lateral roots growing in the upper 80 cm of the soil profile (7). The fungal population in the collar region started to decline following tree death. However, an increase in recovery rate in tap roots at 1.0 and 2.0 m depths following tree death indicated continued activity of the fungus below 1 m. Lack of a parallel increase in fungal recovery rate from the soil surrounding the tap roots suggested that the fungus was growing downward through the root system rather than invading as a front through the soil (Fig. 1). These opposing trends in collar and tap root samples resulted in a two factor interaction of sample depth and age (P = 0.001).

The excavations revealed a consistent pattern of invasion and survival of P. cinnamomi in B. attenuata. The fungus typically infected lateral roots in the upper soil profile, thereby gaining entry to the collar (7). Once the collar had been girdled, the tree died rapidly (T. C. J. Hill, unpublished), and the fungal population near the surface began to decline. At greater depths, invasion of the tap root can continue for some time. After 1 or 2 yr, P. cinnamomi had disappeared from soil and tissue at ground level, but could still be recovered readily from the majority of trees at a depth of 30 cm. After several years, the tree falls over, further accelerating the stump's rate of decay and subsequent fragmentation into small chips. It is likely that the fungus can survive for a considerable time in roots and soil in the moist, white sand below 1 m depth. Similar observations have been made in infected B. grandis Willd. in Eucalyptus marginata Donn. ex Smith forest on lateritic soils further inland. The pathogen was recovered from 92% of collar samples and from up to 98% of soil samples taken from the base of recently dead B. grandis (16,18), while in older, leafless trees the recovery rate from the collar was 17% (16). In the Bassendean Dunes, B. attenuata is one of many tree and large shrub species that would act as a focus of inoculum, and as a refuge from desiccation and microbial attack for the fungus.

Evaluation of glyphosate, metalaxyl, and formaldehyde. Both sites were underlain by Jandakot sand, a podzol characterized by a deep (>5 m), strongly leached A horizon of quartz sand, darkened slightly in the upper meter by organic matter, which overlies a "coffee rock" B horizon formed by the redeposition of iron and organic matter near the water table (2,13). The depth to the winter water table at site 1 was 9 m; at site 2, 6 m. Characteristics of the soils on both sites are given in Table 2. Sites were infested with P. cinnamomi and contained mixed scrub-heath vegetation, the degraded remnants of the original B. attenuata and B. menziesii low woodland.

The method used to test for the presence or absence of *P. cinnamomi* in retrieved pine plugs underestimated the impact of the treatments on fungal populations. With time, as plugs became discolored, decayed, or eaten by termites, the emerging mycelium became sparser and was restricted to discrete regions of the plated surface. At the final assessment, the fungus grew vigorously from only a few of the positively rated plugs. The validity of splitting each plug and plating the exposed surfaces onto selective medium was confirmed by the very

Table 2. Characteristics of soils at treatment sites

Site	Depth (m)	Coarse sand <sup>a</sup> (%)	Fine sand <sup>a</sup> (%)	Silt (%)	Clay (%)	C <sub>org</sub> b (%)
1	0.3	93.2	4.7	0.3	1.9	0.04
2	0.3	91.1	7.4	0.4	1.1	0.07
1	1.3	90.3	8.0	0.1	1.6	< 0.01
2	1.3	83.4	14.7	0.3	1.6	0.01

 $<sup>^{</sup>a}$ Coarse sand = 2.0-0.2 mm; fine sand = 0.2-0.02 mm.

<sup>&</sup>lt;sup>b</sup>Analysed by the Walkley-Black method.

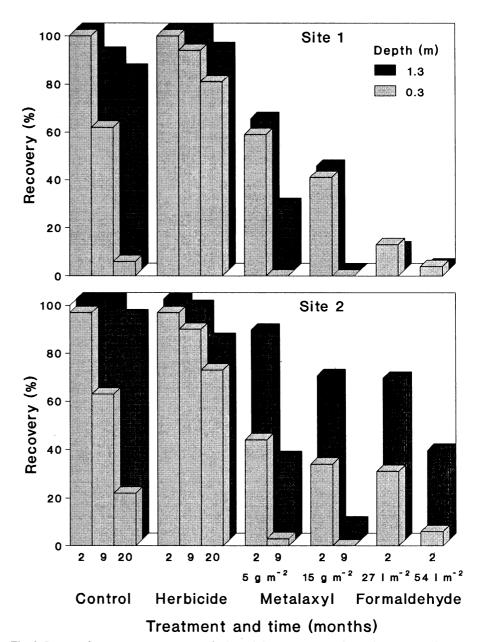


Fig. 2. Impact of treatments on recovery of *Phytophthora cinnamomi* from pine plugs buried at 0.3 m and 1.3 m depth in two sites (n=32 for all histogram bars except formaldehyde, for which n=48) and sampled 2, 9, and 20 mo after treatment. Fungal survival was not assessed in metalaxyl-treated plots after 9 mo, nor in formaldehyde-treated plots after 2 mo. Herbicide, metalaxyl, and formaldehyde (1% v/v) treated plots were completely bare of vegetation.

low recovery rate of the fungus from pieces that were re-split and re-plated after initially having been rated as negative. Of around 600 plugs that were re-tested, only six subsequently produced *P. cinnamomi* colonies. Results of fungal

survival were presented separately for each site due to significant differences (P=0.05) in the efficacy of metalaxyl and formaldehyde in reducing P. cinnamomi survival in plugs buried at 1.3 m.

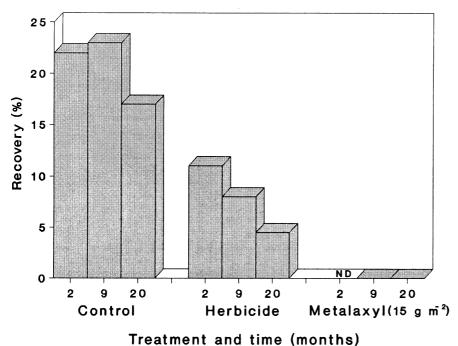


Fig. 3. Effect of treatments on recovery rate of *Phytophthora cinnamomi* from soil core samples (0.1-0.4 m depth), 2, 9, and 20 mo after treatment. Data from two sites is combined (n = 64 for each histogram bar). ND indicates no data.

Glyphosate efficacy. The P. cinnamomi population declined slowly in the completely bare, herbicide-sprayed plots. At 20 mo after treatment, the fungus grew from 77% of plugs at 0.3 m and 87% of plugs at 1.3 m at both sites (Fig. 2). Unexpectedly, P. cinnamomi survived markedly better in plugs at 0.3 m in herbicide-treated plots than in the controls (average of 14%, P = 0.01). This could have been due to the drier soil in control plots during the two summers spanned by the experiment. In control plots, plant water use reduced soil moisture content to 0.6% (n = 40) of dry soil weight at 0.3 m depth in mid-summer, while in herbicide-treated plots it remained at 2.7% (n = 40), comparable with the mid-winter maximum of 3.4%. Herbicide-sprayed plots were insulated by a 10-cm surface layer of completely dry sand that was not in capillary contact with the underlying moist soil. Although fungal survival in plugs at 0.3 m depth may have been enhanced by the death of the vegetation, any seasonal resurgence of fungal activity with the arrival of autumn rains, and a flush of new roots, was prevented. Consequently, the recovery rate of P. cinnamomi from soil samples, always taken in winter or spring, declined steadily to 4.6% in herbicide-treated plots while remaining relatively constant at around 21% in control plots (Fig. 3).

The extended survival of *P. cinna-momi*, particularly at 1.3 m depth, in herbicide and control plots probably is due to the saprophytic growth of the fungus and persistence of its chlamydospores, and is similar to values

found at 30 cm depth in naturally infected B. attenuata stumps (7).

Formaldehyde efficacy. Formaldehyde efficacy differed between sites (Fig. 2). In site 1, P. cinnamomi recovery levels in plots treated with 27 L m<sup>-2</sup> of 1% formaldehyde was reduced to 13 and 9% in plugs at 0.3 and 1.3 m depth, respectively, and to 4 and 2% in plots treated with 54  $L\ m^{-2}$  (Fig. 2). For combined results of both depth categories, the higher dose disinfected significantly more plugs (P = 0.05). In site 2, the impact of formaldehyde was minimal, particularly at a 1.3 m depth where P. cinnamomi was recovered from 67 and 37% of plugs in plots drenched with 27 and 54 L m<sup>-2</sup> of formaldehyde, respectively. Both results were significantly higher than those obtained in site 1 (P = 0.001). Many plugs retrieved from both sites and depths appeared to have escaped contact with the formaldehyde solution or its vapor. Of the 74 P. cinnamomi-positive plugs retrieved from all fumigated plots, 46 showed complete growth; the remainder produced sparse colonies or contained sterilized sectors. It is likely that the unaffected plugs were buried within nonwetting soil pockets that insulated them from the percolating fumigant solution. Dry soil pockets were particularly abundant in the upper 35-50 cm of the profile on both sites, even after the application of heavy rates of soil wetting agent followed by more than 20 cm of rain. Improved fumigation may be achieved by using a smaller volume of a more concentrated solution, and by adding soil-wetting agent to the solution, since the two chemicals do not react.

Inconsistent efficacy of formaldehyde was noted by White and Buczacki (23).

Metalaxyl efficacy. Metalaxyl had a profound, though delayed, effect on P. cinnamomi in both plugs and soil. Recovery rate was reduced dramatically between the first and second sampling times (Fig. 2). At 2 mo after metalaxyl application, P. cinnamomi was recovered from the majority of plugs. However, many colonies exhibited sparse and unusually coralloid hyphae. The effect of both rates of metalaxyl on plugs at 1.3 m in site 2 was less than in site 1, as with formaldehyde-treated plots. Seven months later, P. cinnamomi was almost absent in plots treated with the higher metalaxyl rate (Fig. 2). Of 62 shallow and deep plugs each from both sites, only two 1.3-m-deep plugs from one of the plots in site 2 contained the pathogen. Corresponding recovery rates from control plots were 65 and 95% for plugs at 0.3 and 1.3 m, respectively. For pooled data from both sites, the impact of 15 g (a.i.) m<sup>-2</sup> of metalaxyl was significantly greater than 54 L m<sup>-2</sup> of 1% formaldehyde at a depth of 1.3 m (P =0.005). Phytophthora cinnamomi was also not isolated from either of two batches of 64 soil samples taken 10 and 20 mo after treatment with the heavy metalaxyl dose (Fig. 3). At the same times in control plots, P. cinnamomi was recovered from 23 and 17% of soil samples (n = 64 and P = 0.01 on each occasion). Although both rates of metalaxyl were equally effective against plugs at 0.3 m, the higher dose reduced the average survival rate of the fungus in plugs at 1.3 m from 31 to 3% (P =0.01). All P. cinnamomi-positive plugs at 1.3 m that were exposed to the lower dose did, however, produce minimal colonies, confirming penetration of the fungistat. The very low organic matter content of the Jandakot sand of the study area (0.11% at 0.3 m, and 0.02% at 1.3 m) permitted the deep penetration of metalaxyl (15). A marked decline in the abundance of dry-soil pockets over time, and the possible vapor phase activity of the fungistat (4) may also have enhanced its efficacy.

Similar efficacy of metalaxyl in soil has been achieved in pots and landscape beds (1,3), and in comparable soil in southwest Australia (20), where *P. cinnamomi* was not recovered from any of 20 1-yr-old infected avocado trees, 8 mo after application of 5 g (a.i.) m<sup>-2</sup> of metalaxyl in granular form.

The efficacy of metalaxyl is difficult to explain in light of its fungistatic mode of action. Metalaxyl inhibits growth and especially spore formation in *P. cinnamomi*, but does not kill the fungus (5). However, Malajczuk (11) found a marked interaction between metalaxyl and soil microflora, whereby lysis of *P. cinnamomi* hyphae was greatly accelerated in unsterile soil leachates containing

the fungistat.

Implications for disease management. Sampling of naturally infected B. attenuata stumps, and colonized and buried pine plugs, demonstrated the longevity of P. cinnamomi in this deep sand community. The occurrence of the fungus deep in the profile precludes any practical attempt to eradicate it quickly from infested sites; it had infected around half of the B. attenuata tap roots at 2 m depth, and has been recovered from the water table at 3 and 5 m depth in diseased Banksia woodland (17). Over the long term, however, eradication of the pathogen may be achieved by keeping infested sites and a surrounding buffer area completely bare for an extended period. As the dead hosts decay, and the P. cinnamomi inoculum declines, further spread of the pathogen would be halted by the disintegration of the lateral root network. Species with extensive lateral root systems, such as Macrozamia riedlei Fisch. ex Gaudich., N. floribunda, and E. todtiana may have to be killed within a wider perimeter. Even if complete eradication is not achieved, indefinite containment of a small infested site in an otherwise pristine region is a realistic option. The ongoing maintenance of roads (bare ground) is considered a normal expense, and the limestone roads of the region already appear to be barring the spread of the pathogen in some sites.

When an infected site has the potential to be easily enlarged by human or animal activity, lies close to a drainage line, or immediately threatens rare and restricted endemic flora, metalaxyl could also be used to decontaminate the upper soil layers. Stumps would need to be treated with a fumigant such as 5% formaldehyde. The high cost of chemical treatments would, however, preclude their use in most instances. The total cost of applying metalaxyl (excluding trenching) ranged from \$1.90 to \$4.30  $\text{m}^{-2}$ , depending on rate, while fumigation with formaldehyde costs from \$2.30 to \$3.70  $\mathbf{m}^{-2}$ .

A cheaper, nonchemical alternative for disease control is soil solarization. Recent studies have demonstrated its

potential to eradicate *P. cinnamomi* to a depth of 30 cm (10) or even 45 cm (9) in the soil. The herbicide treatment is very cheap (\$0.10 m<sup>-2</sup> for two applications), and would be simple to maintain with residual, pre-emergent herbicides. Other advantages of using herbicide are that its application only minimally disturbs the site, it can be used to treat large areas without any problems of scale, and the pollution of groundwater does not have to be considered.

The viability of any of these methods is dependent on early detection of infections. Small spot infections expand exponentially to become dieback sites, from which secondary infections are generated. Active protection requires constant monitoring of healthy areas for the first appearance of disease.

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