Injected Treatments for Management of Madrone Canker

Marianne Elliott and Robert L. Edmonds

Abstract. Pacific madrone (Arbutus menziesii) has been experiencing a decline in the Puget Sound area, primarily as a result of a canker disease caused by the fungus Fusicoccum arbuti. Cultural methods such as prevention of stress and wounding are recommended to control canker diseases on trees. In addition to these, injected treatments can be used to protect valuable Pacific madrone trees in urban areas. An experiment testing injectable chemical fungicides and plant activators was performed on Pacific madrone trees inoculated with F. arbuti. There was little correlation between fungicidal activity in culture and canker reduction in the field tests. Two treatments that were effective in minimizing canker growth in inoculated madrones were Arbotect® (Syngenta Crop Protection Inc., Greensboro, NC, U.S.; a triazole fungicide) and BioSerum™ (phosphorous acid). Cankers on wound inoculations were 50% smaller than the control group and no infections occurred on surface-inoculated treatments. Increased callusing was observed on cankers on trees with these treatments and the mode of action for these chemicals is probably stimulation of plant defenses rather than fungicidal action. Phosphorous acid is recommended in addition to cultural methods that improve tree vigor for high-value madrone trees in urban landscapes; however, heavily infected trees that have lost most of their crown will probably not benefit.

Key Words. Arbotect®, Botryosphaeria, canker; Fusicoccum; injectible fungicide; Pacific madrone (Arbutus menziesii); phosphorous acid; plant activator.
Table 1. In vitro testing of fungicides and plant activators. 

<table>
<thead>
<tr>
<th>Fungicide</th>
<th>Active ingredient</th>
<th>Mode of action</th>
<th>Percent inhibition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tebucyl</td>
<td>Tebuconazole</td>
<td>Triazole fungicide, DMI</td>
<td>100</td>
</tr>
<tr>
<td>Fungisol</td>
<td>Debacarb, carbendazim</td>
<td>Benimidazole fungicide, NA</td>
<td>100</td>
</tr>
<tr>
<td>Cleary 3336</td>
<td>Carbenzazim</td>
<td>Benimidazole fungicide, NA</td>
<td>100</td>
</tr>
<tr>
<td>Arbotect&lt;sup&gt;TM&lt;/sup&gt;</td>
<td>Thiabendazole</td>
<td>Benimidazole fungicide, NA</td>
<td>100</td>
</tr>
<tr>
<td>Alamo&lt;sup&gt;®&lt;/sup&gt;</td>
<td>Propiconazole</td>
<td>Triazole derivative fungicide, DMI</td>
<td>100</td>
</tr>
<tr>
<td>Phyton&lt;sup&gt;®&lt;/sup&gt;</td>
<td>Copper</td>
<td>Inorganic fungicide, EI</td>
<td>100</td>
</tr>
<tr>
<td>Cambistat&lt;sup&gt;®&lt;/sup&gt;</td>
<td>Paclorbutrazol</td>
<td>Fungicide and plant activator, DMI</td>
<td>100</td>
</tr>
<tr>
<td>Rovral</td>
<td>Iprodione</td>
<td>Dicarboximide fungicide, NA</td>
<td>75–100</td>
</tr>
<tr>
<td>Bayleton</td>
<td>Triadimefon</td>
<td>Triazole fungicide, DMI</td>
<td>75–100</td>
</tr>
<tr>
<td>NuCop</td>
<td>Copper</td>
<td>Inorganic fungicide, EI</td>
<td>75–100</td>
</tr>
<tr>
<td>Cinnamite</td>
<td>Cinnamaldehyde</td>
<td>Broad spectrum insecticide/miticide/fungicide</td>
<td>75–100</td>
</tr>
<tr>
<td>Kaligreen</td>
<td>Potassium bicarbonate</td>
<td>Inorganic fungicide</td>
<td>&lt;50</td>
</tr>
<tr>
<td>Subdue M axx</td>
<td>Mefanoxam</td>
<td>Acylalanine fungicide, NA</td>
<td>&lt;50</td>
</tr>
<tr>
<td>Aliette</td>
<td>Fosetyl-Al (organic phosphate)</td>
<td>Plant activator, EI</td>
<td>&lt;50</td>
</tr>
<tr>
<td>M-pede</td>
<td>Potassium salts of fatty acids</td>
<td>Inorganic fungicide</td>
<td>&lt;50</td>
</tr>
<tr>
<td>Acligard</td>
<td>Aminazole-s-methyl</td>
<td>Plant activator</td>
<td>&lt;50</td>
</tr>
<tr>
<td>Compass</td>
<td>Strobilurin</td>
<td>Fungicide, ET</td>
<td>&lt;50</td>
</tr>
<tr>
<td>BioSerum&lt;sup&gt;™&lt;/sup&gt;</td>
<td>Phosphorous acid</td>
<td>Plant activator</td>
<td>&lt;50</td>
</tr>
<tr>
<td>Control</td>
<td>N/A</td>
<td>N/A</td>
<td>0</td>
</tr>
</tbody>
</table>

<sup>a</sup>Results were based on fungal growth after 7 days at 25°C (77°F). Percent radial growth inhibition was calculated as the difference from control growth.

<sup>b</sup>Chemicals used in the field tests.

DMI = sterol demethylation inhibitor; NA = nucleic acid and protein synthesis inhibitor; ET = electron transport disruptor; EI = nonspecific enzyme inhibitor; N/A = not applicable.

...der pressure, and infusion, in which a large volume of dilute solution is fed into the tree through gravity and taken up in the transpiration stream. Treatments should be performed on a day when the tree is actively transpiring and after a period of heavy rainfall for best results (Stipes 1988). These methods do not eradicate the pathogenic fungus, but reduce symptom expression and inoculum density so that the tree can be healthy enough to defend itself. Injected fungicide or plant activator treatments in combination with cultural methods to improve tree vigor have the most likelihood of success. Injection treatments on DED are advised when there is no more than 5% to 10% of the crown affected by disease. In general, injection treatments are not recommended for trees with severe disease symptoms (Lanier 1988). Trees of reduced vigor are less likely to recover from injuries sustained during injection. There are no reports of injected treatments being used to manage Botryosphaeria diseases on trees.

The best treatments for madrone canker are ones that will improve overall tree vigor and include cultural methods such as sanitation, prevention of wounding, and improving soil water retention. Additional protection can be used in the form of chemical treatments, especially on high-value trees. Systemic fungicides and plant activators will give additional disease resistance, but are costly and can be damaging in some situations.

The objective of this study was to evaluate the ability of several injected chemicals to reduce damage to Pacific madrone caused by the canker pathogen F. arbuti.

**METHODS**

**In Vitro Testing of Fungicides**

Eighteen fungicides and plant activators were tested for efficacy against F. arbuti in culture (see Table 1 for list; mode of action is indicated). Two hundred parts per million active ingredient of each chemical was added to 2% malt extract agar. Controls were plates with 2% malt extract agar and no added chemical. Nine isolates of F. arbuti collected from the range of Pacific madrone, from southern California to Washington State, were tested using three replicates of each fungal isolate. Plates were inoculated from plugs taken from active cultures and incubated at 25°C (77°F; the optimal temperature for F. arbuti radial growth) and fungal radial growth was measured at intervals of 7 days for 1 month and expressed as millimeters per day.

**Results**

Fungal radial growth was calculated as the difference from control growth. The best treatments for madrone canker are ones that will improve overall tree vigor and include cultural methods such as sanitation, prevention of wounding, and improving soil water retention. Additional protection can be used in the form of chemical treatments, especially on high-value trees. Systemic fungicides and plant activators will give additional disease resistance, but are costly and can be damaging in some situations.

The objective of this study was to evaluate the ability of several injected chemicals to reduce damage to Pacific madrone caused by the canker pathogen F. arbuti.

**Field Tests of Systemic Treatments**

The field tests took place on Lopez Island, Washington. Forests in this region are drier than those on the mainland as a result of the influence of the Olympic Mountains and Vancouver Island situated southwest and west northwest of the San Juan Islands. These land masses create a “rain shadow” effect that produces less rainfall in the islands than the rest of northern Puget Sound. At the nearest weather station at Olga on Orcas Island, the average high temperature in July is 21°C (69.8°F), and the average low in January is 2°C (35.6°F) (WRCC 2006). Winter temperatures are usually mild, except when cold, arctic air funnels down the Fraser River Valley from Canada and drastically lowers temperatures. Winds may also have a drying effect, pulling moisture from vegetation and surface water, and creating drought stress during low rainfall periods.

One hundred twenty-eight Pacific madrone trees were selected for field tests of the chemical treatments. These trees were located in a forested area with Douglas fir (Pseudotsuga menziesii) canopy dominants. The madrones formed a layer under the Douglas fir and many showed signs of competition for space and light. Most of the trees were intermediate or suppressed crown classes and growing at the edge or interior of the stand. The stand was considered to be intermediate in moisture conditions between dry and moist woodland. Soils are deeper on this site (San Juan County Health and Community Services 2000) than in other areas of the San Juan Islands and have a thick duff layer. Typical shrubs were salal (Gaultheria shallon), Oregon grape...
(Mahonia nervosa), ocean spray (Holodiscus discolor), and little wild rose (Rosa gymnocarpa).

Many of the trees in the stand were infected with F. arbuti in varying degrees of severity. Lopez Island is in an area severely affected by Pacific madrone decline (USFS 2003) and cankers on some trees were at least 15 years old as estimated by the number of callus layers. These trees on Lopez Island were good candidates for treatment because Fusicoccum is more aggressive on stressed trees and any treatment that was successful on them could have a good possibility of being effective on urban trees taking into account environmental factors that may influence disease development.

Each tree was evaluated and data were collected on diameter at breast height (dbh) and percent dieback, crown, and live foliage (Table 2) and randomly placed into one of eight treatment groups with 16 trees per group. The chemical treatments selected for the field tests were Alamo, Arbotect®, BioSerum™, Cambi-stat, Fungisol, Phyton, Tebuject, and a water control (see list in Table 1). These chemicals were selected based on their behavior in vitro and also to represent different chemical modes of action. All chemicals except BioSerum™ caused 100% inhibition of mycelial growth in vitro, but BioSerum™ was selected for use in field tests because of its plant activator properties. Injection treatments were applied in Summer 2002 according to the label instructions for elm or oak.

The test madrones were inoculated in September 2002 with a single isolate of F. arbuti taken from a Pacific madrone tree in Port Townsend, Washington. This isolate was chosen because it was close to the field site in geographic origin. Three wounds were made on branches of each tree and a small amount of fungal inoculum (approximately 3 mm³ [0.12 in³]) from a Pacific madrone tree in Santa Cruz Co., CA 43.5 ab 79.3 bc 50.5 ab −5.1 a 74.4 ab 0.47 ab

7. Nevada Co., CA 50.1 ab 71.7 ab 45.7 ab 22.6 ab 69.0 ab 4.2 ab

8. Santa Cruz Co., CA 43.5 ab 79.3 bc 50.5 ab −5.1 a 74.4 ab 0.47 ab

9. Santa Cruz Co., CA 39.4 ab 77.6 ab 31.4 a 20.8 bc 69.7 a −6.1 a

Isolate Actigard Bayleton Compass Kaligreen Nucop Subdue Maxx

1. King Co., WA 44.8 ab 82 c 50.2 ab −5.1 ab 78.4 ab 3.8 ab
2. King Co., WA 51.1 ab nt 45.7 ab 22.6 c 80.0 ab 2.2 ab
3. King Co., WA 54.8 b 100 d 64.8 b 15.4 abc 86.9 b 15.1 b
4. Vashon, WA 27.8 a 78.9 bc ab 47.5 ab 18.9 bc 75.8 ab 8.0 ab
5. Vashon, WA 45.9 ab 79.9 b 40.2 a 15.5 abc 75.6 ab 0.7 ab
6. Trinity Co., CA 42.6 ab 100 d 50.9 ab −1.4 ab 76.7 ab 4.3 ab
7. Nevada Co., CA 50.1 ab 71.7 a 56.3 a 22.9 c 71.4 a 7.2 ab
8. Santa Cruz Co., CA 43.5 ab 79.3 bc 50.5 ab −5.1 a 74.4 ab 0.47 ab
9. Santa Cruz Co., CA 39.4 ab 77.6 ab 31.4 a 20.8 bc 69.7 a −6.1 a

Isolated growth was measured in millimeters per day at 25°C (77°F). Negative values indicate that the fungicide stimulated fungal growth relative to the control. Isolates with the same letter in each fungicide column are not different in percent inhibition (P = 0.05, Dunnett’s T3 test).
Table 4. Effect of injected treatments of fungicides and plant activators on Pacific madrone canker growth.\(^a\)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>2003</th>
<th>2004</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>124.8 (19.35)</td>
<td>192.42 (29.83)</td>
</tr>
<tr>
<td>Alamo</td>
<td>88.43 (13.71)</td>
<td>113.97 (17.67)</td>
</tr>
<tr>
<td>Arbotect(^b)</td>
<td>83.13 (12.89)</td>
<td>93.61 (14.51)</td>
</tr>
<tr>
<td>BioSerum(^m)</td>
<td>48.81 (7.57)</td>
<td>62.72 (9.72)</td>
</tr>
<tr>
<td>Cambistat</td>
<td>102.83 (15.94)</td>
<td>165.69 (25.68)</td>
</tr>
<tr>
<td>Fungisol</td>
<td>92.54 (14.34)</td>
<td>177.77 (27.55)</td>
</tr>
<tr>
<td>Phyton</td>
<td>100.03 (15.50)</td>
<td>117.21 (18.17)</td>
</tr>
<tr>
<td>Tebuject</td>
<td>90.32 (14.00)</td>
<td>115.76 (17.94)</td>
</tr>
</tbody>
</table>

\(^a\)Mean area of inoculated canker in centimeters squared (inches squared) is given for each treatment during a 2-year period measured in 2003 and 2004. Treatments with the same letter in year column are not significantly different at \(P = 0.05\) (Dunnett’s T3 test).

which surface inoculations developed into cankers were Alamo (7%), Fungisol (13%), Phyton (21%), and control (17%).

Although the differences were not significant, concentrations of phenolic defense chemicals were higher in trees treated with phosphorous acid, whereas the chemical itself was ineffective as a fungicide when tested in culture. The fungicide treatment groups were not significantly different when further samples were collected on the same age foliage in 2004 (1 year after treatment). Levels of foliar phenolics were highest in the Arbotect\(^b\) and BioSerum\(^m\) treatments, but these decreased after 1 year (Table 5). Values for other treatments increased after 1 year in the same foliage. In new foliage 1 year after treatment, the difference between groups was significant (\(P = 0.011\), Kruskal-Wallis test) with the control and Cambistat treatments having the lowest amount of foliar phenolics and Phyton the highest.

No relationship was found between tree dbh and canker area for individual treatments for both years. A similar result was obtained for the relationship between total foliage and canker area, except in the Arbotect\(^b\) treatment, in which trees with more foliage had smaller cankers (\(r^2 = 0.55, P = 0.04\) for 2003, \(r^2 = 0.64, P = 0.02\) for 2004). There was more of a difference in canker size between treatments 1 year after treatment, and treatment effects seemed more important than the initial health of the tree.

**DISCUSSION**

**In Vitro Testing of Fungicides**

Because chemical fungicides have not been used with *F. arbuti*, the concentration of 200 ppm active ingredient was chosen based on in vitro screening of fungicides for use against *Botrytis cinerea*, in which the 200 ppm dose was found to be suitable for separating effective and ineffective fungicides (W. Littke, pers. comm.). Triazoles and benzimidazoles were most effective as fungicides against *F. arbuti* in culture. Li et al. (1995) using in vitro testing of fungicides against *Botryosphaeria dothidea* (*Fusicoecum aesculi*) found the most suppression of mycelial growth in plates containing carbendazim and thiophanate-methyl at concentrations of 50 and 100 ppm. These concentrations were lower than the 200 ppm active ingredient concentration used in this study. Fosetyl-Al and phosphorous acid were both inhibitory to *Phytophthora* spp. in culture (Fenn and Coffey 1983) at concentrations ranging from 69 to 552 ppm, but we found they were not inhibitory to *F. arbuti* at 200 ppm. Fenn and Coffey (1983) report that these chemicals had low activity against a range of non-Oomycete pathogens so this result is not surprising.

Copper was more effective in inhibiting fungal growth than potassium, but after a period of time in culture, the fungus was observed to accumulate copper from the medium and isolate it. After that, the mycelium was able to grow unchecked. The melanized hyphae of *Fusicoecum* protected the mycelium from lysis by potassium salts. These fungicides are commonly used to control powdery mildews (*Fusicoecum* et al. 2002) whose hyphae may be more easily disrupted. The response to cinnamaldehyde was extremely variable, perhaps as a result of the volatility of the chemical.

There was variability in tolerance to some fungicides among the nine isolates of *F. arbuti* tested. All were completely inhibited by 200 ppm of the triazole fungicides tebuconazole (Tebuject) and propiconazole (Alamo). In a study of sensitivity of *B. dothidea* to tebuconazole, M a et al. (2001b) found that sensitivity to tebuconazole correlated significantly with sensitivity to propiconazole. Fungicide concentrations ranged from 0.0625 to 1.0 ppm, much lower than the 200 ppm used in this study. M a et al. (2001c) found that some isolates of *B. dothidea* developed resistance to iprodione (Rovral) in vitro and remained resistant when treated with up to 500 ppm of the fungicide. There was a range of tolerance to this chemical among isolates of *F. arbuti* at 200 ppm, but the differences were not statistically significant. Triazole and benzimidazole fungicides were the most effective against *F. arbuti* in vitro and we anticipated that this would translate to reduction of fungal growth in established infections. Inhibition of radial growth by chemicals with fungicidal activity can be an estimator of fungicidal activity in infected host canker tissue, and this was confirmed by some of the field results. However, not all the fungicides that performed well in culture did so in the field and vice versa. How well these chemicals behave in prevention of new infections was not tested in this study and should be done by measuring inhibition of spore germination.

**Field Tests of Systemic Treatments**

Because we did not want to introduce new genotypes of *F. arbuti* to the field site, we chose a local isolate of *F. arbuti* to be used in the field tests. There were no significant differences in sensitivity of isolates from the Puget Sound area to fungicides used in the field tests in vitro, so this isolate could be considered representative of the region. However, a more complete test should include isolates of *F. arbuti* from the entire range of Pacific madrone to fully account for genetic variability.

---

©2008 International Society of Arboriculture
**Fusicoccum arbuti** appears to be a wound-invading pathogen on woody tissue because 100% of wound inoculations on all trees developed cankers, whereas only 3% of surface inoculations did. No cankers developed from bark surface inoculations in the Arbotect™, BioSerum™, Cambistat, and Tebucbit treatments. Perhaps defenses in the bark were stimulated, preventing fungal colonization.

The benzimidazole fungicides, with the exception of Arbotect™, performed less well in the field than in vitro. BioSerum™ (phosphorous acid) was the most effective fungicide in field tests. This is probably attributable to plant defenses being activated and not from fungicidal activity of the chemical. BioSerum™ and a similar product, A liette (Fosetyl-AI), were not inhibitory in culture. The previous in vitro fungicide results suggest the effectiveness of phosphorous acid is the result of stimulating plant defenses rather than direct fungicidal activity.

Treatment with phosphorous acid holds the most promise for madrones in disease prevention and in treating preexisting cankers. Because wounding and disease cause a defense response in trees (Percival 2001; Krokene et al. 2003), induced defenses already present resulting from active cankers on the tree before treatment with BioSerum™ were increased after treatment. Phosphorous acid, the active ingredient in BioSerum™, has been demonstrated to have a stimulatory effect on host defense responses to infection by *Phytophthora* spp. (Fenn and Coffey 1983).

Arbotect™ performed almost as well as BioSerum™ and a similar (induced) level of phenolic defense chemicals was seen in the foliage, but decreased in the second year after treatment. Cankers on these two treatments appeared to be more callused than on other treatments. Arbotect™ may behave as a plant activator in addition to having fungicidal properties. Perhaps production of phenolic defense chemicals is less important than growth of callus tissue as a mode of plant defense initiated by these two chemical treatments, although increased foliar phenolics may prevent colonization by fungal pathogens.

All of these fungicides, except for benomyl, are sterol demethylation inhibitors and prevent formation of ergosterol in fungi. However, effectiveness of these fungicides may be the result of their behavior in the host rather than fungicidal activity. Sterol-inhibiting fungicides are known to change the balance of plant hormones and reduce transpiration rates in some crops (Lonsdale and Kotze 1993). This confers drought tolerance on the plant and possible resistance to canker fungi such as *Botryosphaeria* spp., that are pathogenic when the host is under water stress. New infections are prevented during times when stomates are closed.

**Implications for Arboriculture**

Treating Pacific madrone trees with phosphorous acid (BioSerum™) or Arbotect™ will stimulate their defenses and reduce the severity of cankers caused by *F. arbuti*. These treatments also will prevent some new infections through increased defense chemicals in the foliage. The treatment effect lasts at least 1 year. These treatments may be effective on diseases caused by *Botryosphaeria* spp. in other hosts in addition to Pacific madrone.

**Acknowledgments.** We thank Fred Ellis of Island Foresters and Debbie Hayward of Vulcan Corp. for assistance with fieldwork and chemical applications and Will Littke, Weyerhaeuser Corporation, for help with laboratory work.

**LITERATURE CITED**


Marianne Elliott (corresponding author)
Canadian Forest Service
Pacific Forestry Centre
506 West Burnside Road
Victoria, BC, Canada V8Z 1M5
mellott@nrcan.gc.ca

Robert L. Edmonds
University of Washington
College of Forest Resources
Box 352100
Seattle, WA 98195, U.S.

Résumé. L’arbusoir de M enziesi (Arbutus menziesii) a souffert de dépérissement dans la région du détroit du Puget en raison principale du chancre causé par le champignon pathogène Fusicoccum arbuti. Les méthodes culturelles telles que la prévention des stress et la chirurgie sont des traitements recommandés pour les arbres atteints par le chancre. Des traitements par injection peuvent aussi être utilisés pour protéger l’arbusoir de M enziesi dans les zones urbaines. Une expérience mettant à l’essai l’injection de fongicides chimiques et de d’activateurs végétaux a été menée sur des arbusiers de M enziesi inoculés avec F. arbuti. Dix-huit fongicides ont été testés en culture et sept parmi ces derniers lors de tests sur le terrain. Il y avait une faible corrélation entre l’activité fungique en milieu culturel et la réduction des chancres dans les tests sur le terrain. Deux traitements qui ont été efficaces pour minimiser la croissance des chancres sur des arbusoirs inoculés étaient l’Arbotect® (un fongicide à base de triazole) et le BioSerum™ (acide phosphorique). Les chancres inoculés sur des blessures étaient 50% plus petits que ceux du groupe témoing et aucune infection n’est survenue sur les sujets inoculés en surface. La formation d’un cal plus important a été observée autour des chancres sur des arbres avec ces traitements et le mode d’action de ces substances chimiques était probablement plus une stimulation des défenses de la plante qu’une action directe du fongicide. L’acide phosphorique est recommandé en sus aux méthodes culturelles afin d’améliorer la vigueur de l’arbre, et ce pour les arbusoirs de M enziesi de grande valeur dans les aménagements en milieu urbain.


Resumen. El madroño del pacific (Arbutus menziesii) ha estado sufriendo declinación en el área de Puget Sound, primariamente debido a la enfermedad de cancro causada por el hongo Fusicoccum arbuti. Los métodos culturales tales como prevención del estrés y manejo de cavi-
dades son tratamientos recomendados para enfermedades de cancro en los árboles. Los tratamientos con inyecciones también pueden ser usados para proteger madroños valiosos en áreas urbanas. Se realizó un experimento probando fungicidas químicos inyectables y activadores vegetales en árboles de madroño inoculados con F. arbuti. Fueron probados 18 fungicidas en cultivo, siete de los cuales fueron usados en pruebas de campo. An Bäumen mit dieser Behandlung wurde vermehrt Kallusbildung beobachtet und die aktive Komponente in diesen Chemikalien ist vermutlich eher eine Stimulans der körper-eigenen Abwehr als eine fungizide Wirkung. Phosphorsäure wird zusätz-
liz zu kulturellen Methoden empfohlen, um die Vitalität wertvoller Bäume in urbanen Gegenden zu verbessern.

©2008 International Society of Arboriculture