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Efficacy of Three Macroinfused Fungicides to Control Laurel Wilt on Avocado in Martin and Brevard Counties

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Laurel wilt, a lethal disease of trees in the Lauraceae, is caused by *Raffaelea lauricola* (*Raf*) which has several ambrosia beetle vectors. Three rates each of macroinfused thiabendazole (Arbotect® 20-S), propiconazole (PropiconazolePro®), and tebuconazole (Teb #1—proprietary formula, Rainbow Treecare) were tested for efficacy against the disease on avocado trees in Martin and Brevard Counties. Seven trees were not treated with fungicide, and five to six were infused with Arbotect® (1.0, 2.0 or 3.0 oz. of product per inch trunk dia.), or PropiconazolePro® or tebuconazole (0.7, 1.0, and 1.4 oz. per inch trunk dia.) on 12–13 July 2011. On 13–14 October 2011, all trees were inoculated with the pathogen, and 60, 239, and 558 days after inoculation (dai) laurel wilt development was rated. Laurel wilt developed to a greater extent on trees treated with either tebuconazole or Arbotect®, 60 dai, than those treated with PropiconazolePro® (no symptoms developed at any rate of this fungicide). However, by 239 dai a large proportion of the treated trees had developed laurel wilt symptoms, regardless of the fungicide that was used (mean incidences of 83%, 89%, and 94% for, respectively, PropiconazolePro®, Teb#1, and Arbotect®). By 239 and 558 dai, disease severities and the recovery of the pathogen were lower in trees treated with PropiconazolePro® or tebuconazole, compared to Arbotect®. Although lower disease severities developed in the former trees, disease variability among trees within treatments suggests that other factors (e.g., cultivar and rootstock) may have influenced the results. Current recommendations for managing this disease are discussed.

Laurel wilt is a lethal disease of trees in the Lauraceae caused by *Raffaelea lauricola* (*Raf*) (Fraedrich et al., 2008). The pathogen is a symbiont of the redbay ambrosia beetle (*Xyleborus glabratus*) (RAB). Indigenous to subtropical areas of Asia (i.e., India, Taiwan, China, Japan, and Myanmar) this disease-insect complex was introduced into the United States through Port Wentworth, GA, in 2002. By 2004 it was clear that laurel wilt was responsible for the death of native redbay (*Persea borbonia*) trees in southeastern Georgia. The RAB vector spread laurel wilt through non-agricultural areas to the north, south and west, and by 2005 it was detected in Duval County, Florida, probably as a result of movement of RAB-infested wood from Georgia (Mayfield and Thomas, 2006). Currently, laurel wilt has been detected in North and South Carolina, Georgia, Florida, Alabama, Mississippi, Louisiana, and Texas (Anonymous, 2014; Bates et al., 2015).

In Florida, the spread of RAB in the wild and human transport of wood products continued, and by 2006, 2007, and 2008 laurel wilt was detected in the mid-southeastern Florida coastal counties of Indian River, Brevard, and Martin Counties, respectively (Gardner et al., 2015). Currently laurel wilt has been detected in 59 of Florida's 67 counties, including Miami-Dade County where 98% of Florida's commercial avocado production resides.

Preliminary tests conducted in north Florida showed that containerized avocado trees were attacked by RAB and succumbed to laurel wilt (Mayfield et al., 2008). Research into the biology and control of laurel wilt and RAB began in earnest in 2006 and by 2011 three fungicides, propiconazole, tebuconazole, and thiabendazole were identified as potential prophylactic treatments to prevent laurel wilt in avocado trees (Ploetz et al., 2011). However, since laurel wilt was not present in Miami-Dade County at that time, a field trial to determine the efficacy of these fungicides was initiated in small avocado plantings in Martin and Brevard (Merritt Island) Counties (where the disease was present).

Materials and Methods

During Fall 2010, 60 mature avocado (*Persea americana*) trees of varied pedigree were identified in commercial groves in Martin County (18 trees) and Merritt Island, Brevard County (42 trees). In these areas, laurel wilt was widespread on native redbay trees and was evident on some avocado trees. The racial background of the identified avocado trees included West Indian ('Pollock'), Mexican ('Zutano'), Guatemalan x Mexican ('Winter Mexican'), Guatemalan x West Indian ('Lula', 'Choquette', 'Monroe', 'Hall', 'Day', and 'Marcus Pumpkin') and 'Brogdon', a complex hybrid (Schnell et al., 2003). However, the genetic background was not known for about half the trees. Tree health was noted during Spring 2011, prior to the start of the investigation with many of the avocado trees in Martin County exhibiting symptoms of cold damage (i.e., stem dieback and leaf drop) and

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Table 1. Fungicide active ingredient, percent active ingredient, brand name, and rate per inch mean trunk diameter.

Fungicide active ingredient	Active ingredient (%)	Brand name and manufacturer	Rate per inch of mean trunk diameter (oz)
Propiconazole	14.3	PropiconazolePro®, Micro Flo Co., Memphis, TN	0.7, 1.0, 1.4[z]
Tebuconazole	16.0	Teb #1 – proprietary formula Rainbow Tree Science, Minnetonka, MN	0.7, 1.0, 1.4[z]
Thiabendazole	26.6	Arbotect® 20-S, Syngenta, Greensboro, NC	1.0, 2.0, 3.0[y]

[y]1.0, 2.0 and 3.0 ounces equals 29.5, 59.1, and 88.7 mL, respectively.

[z]0.7, 1.0, and 1.4 ounces equals 20, 30, and 40 mL, respectively.

Table 2. Fungicide efficacy against laurel wilt on mature, field grown avocado trees.

		Canopy with laurel wilt symptoms								
		Percentage of trees showing laurel wilt symptoms			Mean percentage of a given tree canopy with laurel wilt symptoms (%)			Number and percentage of trees from which *Raf* was recovered[y]		
Treatment	Fungicide rate (oz)[z]	60 dai[x]	239 dai[x]	558 dai[x]	60 dai[x]	239 dai[x]	558 dai[x]	60 dai[x]	239 dai[x]	558 dai[x]
Non-treated	---	86	100	100	66	80	75	5/6 (83%)	6/6 (100%)	nd
Propiconazole	0.7	0	83	67	0	20	31	0/6 (0%)	0/6 (0%)	1/6 (17%)
	1.0	0	100	83	0	33	43	0/6 (0%)	1/6 (17%)	nd
	1.7	0	67	50	0	19	56	0/6 (0%)	1/6 (17%)	nd
Means/totals	**---**	**0**	**83**	**67**	**0**	**24**	**43**	**0/18 (0%)**	**2/18 (11%)**	**1/6 (17%)**
Tebuconazole	0.7	33	83	67	4	13	41	2/6 (33%)	3/6 (50%)	4/6 (67%)
	1.0	0	83	67	0	20	52	0/6 (0%)	0/6 (0%)	2/6 (33%)
	1.7	17	100	67	16	25	26	1/6 (17%)	1/6 (17%)	3/6 (50%)
Means/totals	**---**	**17**	**89**	**67**	**7**	**19**	**40**	**3/18 (17%)**	**4/18 (22%)**	**9/18 (50%)**
Thiabendazole	1.0	60	80	60	14	42	61	2/6 (33%)	3/6 (50%)	4/6 (67%)
	2.0	50	100	50	19	28	62	2/6 (33%)	3/6 (50%)	4/6 (67%)
	3.0	67	100	67	30	68	73	4/6 (67%)	5/6 (83%)	5/6 (83%)
Means/totals	**---**	**59**	**93**	**59**	**21**	**46**	**65**	**8/18 (44%)**	**11/18 (61%)**	**13/18 (72%)**

[z]Volume of product per inch of trunk diameter.

[y]Number of trees from which *Raf* was recovered / number of trees assayed for the pathogen; nd = not determined; (%) = percentage of treated trees that were positive for *Raf*.

[x]60 days after inoculation (dai) = Dec. 12 and 13, 2011; 239 dai = June 7 and 8, 2012; 558 dai = April 24–26, 2013.

salinity stress (e.g., marginal leaf necrosis, chlorosis). By the time fungicide treatments were applied in July, most had healthy new growth. Similarly, some trees in Merritt Island showed signs of salinity stress, but in general trees were in moderately good to good condition by summer.

The efficacy of three fungicide to prevent laurel wilt, propiconazole (PropiconazolePro®), tebuconazole (Teb #1—proprietary formula), and thiabendazole (Arbotect®) were investigated at three rates (Table 1). Propiconazole and tebuconazole rates were 0.7, 1.0, and 1.7 oz per inch of trunk diameter. Thiabendazole rates were 1.0, 2.0, and 3.0 oz per inch of trunk diameter. Either five or six trees were assigned one fungicide active ingredient and rate and macroinfused over a four-day period (11–14 July 2011) (Table 1). Seven trees were not infused with fungicide but subsequently inoculated with the pathogen (*Raf*) and served as non-treated controls. Many trees had multiple trunks and individual trunk diameters ranged from 3.5 inches to 12.5 inches; trunk diameter means and fungicide rates per inch of trunk diameter were used to calculate the amount of fungicide infused per tree. Each fungicide was mixed with approximately two gallons of water. The Teb#1 formulation that was used tended to crystalize, which made its infusion difficult. The Rainbow Treecare™ (Minnetonka, MN) low volume macro pump kit (Model 5305) was used to infuse trees with fungicide. Per Rainbow instructions, trees were infused in root flares under about 15 psi pressure. The time for trees to absorb the fungicide ranged from 20 min. to about 12 h. Approximately 97 days after fungicide infusion (13–15 Oct. 2011) trees were inoculated at 3 to 17 sites (depending upon tree size and number of trunks) along their trunks and major limbs with 100 μL (containing 100,000 conidia) per site with *Raf*. The inoculum was placed into the sapwood of each tree by drilling a 3-inch-deep hole with a 15/64 inch diameter drill bit and micro-pipetting the inoculum into the hole. Inoculation sites were plugged with putty after loading to seal the hole. Seven trees were not infused with fungicide but inoculated with *Raf* and served as inoculated controls.

The percentage of trees showing symptoms of laurel wilt (e.g., wilting, desiccated leaves retained on dead stems, and limb dieback) and the mean percentage of a given tree canopy with laurel wilt symptoms was recorded: 60 (12–13 Dec. 2011), 237 (7–8 June 2012) and 558 (24–26 Apr. 2013) days after inoculation (dai) with *Raf*. Chips of xylem sapwood were removed from some of the symptomatic trees to confirm presence of the pathogen (Ploetz et al., 2012).

Results and Discussion

Sixty days after inoculation with *Raf*, 86% of the non-treated control trees (i.e., not treated with fungicide) developed laurel wilt symptoms (Table 2). By 239 dai, all of these trees were symptomatic and positive for *Raf*. By 60, 239 and 558 dai the mean percentage of canopy with laurel wilt symptoms for nontreated trees was 66%, 80%, and 75%, respectively.

At 60 dai, none of the trees treated with propiconazole had laurel wilt symptoms (Table 2). In contrast, symptoms of laurel wilt were evident on 33% and 17% of those trees infused with tebuconazole at the 0.7 oz and 1.7 oz rate, respectively and 50% to 67% of those trees treated with thiabendazole at the 1.0, 2.0. and 3.0 oz. rate, respectively. The mean percentage of tree canopy symptoms was low (7% and 21%) for tebuconazole and thiabendazole infused trees, respectively. By 239 dai, the percentage of trees showing laurel wilt symptoms (disease incidence) was 83%, 89%, and 93% for all three rates of propiconazole, tebuconazole, and thiabendazole infused trees, respectively (Table 2). However, the mean percentage of canopy with symptoms of laurel wilt remained relatively low (disease severity) at 24%, 19%, and 46% for all three rates of propiconazole, tebuconazole, and thiabendazole infused trees, respectively. By 558 dai, disease severity increased about 20% for all treatments.

As expected, *Raf* was recovered from relatively few trees that were treated with any of the fungicides (by 558 dai, 1/6, 9/18, and 13/18 of the assayed trees that were treated with propiconazole, tebuconazole, and thiabendazole, respectively) (Table 2). Prior work demonstrated a dramatic reduction of viable pathogen in treated, inoculated trees (Ploetz unpublished). Symptom development is a more reliable indicator of the efficacy of fungicides that are used to manage this disease.

At the rates of fungicide and formulations investigated, the PropiconazolePro® was more efficacious than the Teb#1 or Arbotect® 20-S over an 18 month period. Currently, another formulation of propiconazole, Tilt® has an emergency use (Section 18) label for prevention of laurel wilt in commercial avocado trees. Tilt® is being used primarily to prevent the spread of the laurel wilt pathogen among root grafted avocado trees in commercial groves.

The current recommendations for control of laurel wilt include:

1. Early detection of laurel wilt through aerial surveys and ground based scouting.
2. Sanitation: Immediate uprooting and destruction of all above and below ground parts of affected trees by chipping and/or burning.
3. Treatment of chips with contact chemical (e.g., Malathion or Danitol) or bio-insecticides (e.g., BotaniGard or Mycotrol).
4. Two, tree directed chemical or bio-insecticide applications in the area of laurel wilt affected trees.
5. Infusion or injection with Tilt® (propiconazole) of two to three adjacent healthy avocado trees in all directions of laurel wilt affected trees; called spot treatment.
6. Optionally, sever root systems of laurel wilt and adjacent healthy avocado trees by trenching a perimeter.
7. Continued scouting and immediate application of recommendations 1 through 6.

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