

Effectiveness of Different Fungicides Applied Preharvest at Reducing Postharvest Decay of Fresh Florida Citrus

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Annual studies have been conducted since 1999 to evaluate the effectiveness of preharvest applied compounds in reducing postharvest decay of many types of fresh citrus fruit (*Citrus* spp.). Commercially mature fruit were usually harvested at two different times after the compounds were applied, degreened when necessary, washed, waxed (without fungicide), and then stored at 50 °F (10.0 °C) with about 90% relative humidity. Compared to control (unsprayed) fruit, preharvest application of benomyl (Benlate) or thiophanate-methyl (Topsin) most often resulted in significantly ($P < 0.05$) more healthy citrus fruit and less decay from stem-end rot (SER; primarily *Lasiodiplodia theobromae* or *Phomopsis citri*) after storage, often reducing total decay by about half. Pyraclostrobin (Headline), pyraclostrobin + boscalid (Pristine), and phosphorous acid (Phostrol) only occasionally reduced decay from SER or anthracnose (*Colletotrichum gloeosporioides*). None of the other tested materials reduced postharvest decay or increased the number of healthy fruit after storage.

Control of postharvest decay is a major concern for fresh fruit growers and shippers. Postharvest decay losses are not only costly, but also diminish domestic and export consumer confidence in product quality. Unlike Mediterranean climates where penicillium (*Penicillium* spp.) mold is the predominant cause of citrus decay, stem-end rot (SER; primarily *Lasiodiplodia theobromae* or *Phomopsis citri*) often causes the greatest postharvest losses in fresh Florida citrus (Brown and Albrigo, 1970). Decay due to anthracnose (*Colletotrichum gloeosporioides*) can also be severe, especially in early-season tangerines (Brown, 1978). Postharvest fungicides are used in most commercial packinghouses to control these and other postharvest decay organisms. However, preharvest application of benomyl or thiophanate-methyl, with their ability to reduce fruit decay through harvest, packing and marketing operations, provides another important management option for growers and shippers (Brown and Albrigo, 1970, 1972; Ritenour and Timmer, 2007; Ritenour et al., 2004; Zhang and Timmer, 2007). Preharvest use of these fungicides decreases postharvest decay during times of high decay pressure and may allow for the use of fewer postharvest fungicides. Furthermore, they can greatly retard decay development between harvest and packing operations which is especially important when warm weather retards natural color development and degreening of the fruit with ethylene is required. Ethylene exposure and the warm [up to 85 °F (29.4 °C)], humid (~95% relative humidity) conditions during degreening increase the incidence of diplodia SER and anthracnose (Brown and Barmore, 1976; Grierson and Newhall, 1955). While fungicide drenches before degreening are common, early season fruit peel may be injured by the drench chemicals (Ritenour and Dou, 2000).

Effective preharvest fungicides with continuing postharvest decay control is an important management option for the fresh

citrus industry. The manufacture and sale of benomyl (Benlate; Dupont, Wilmington, DE) was discontinued in late 2001. Of the remaining available products, only thiophanate-methyl is effective at reducing postharvest SER development under Florida conditions (Ritenour et al., 2004; Zhang and Timmer, 2007). With the constant threats of losing currently available fungicides and development of fungicide resistance in decay-causing organisms (McDonald et al., 1979), there is also a need to expand and diversify decay control options for citrus. The current paper summarizes results from studies conducted between 1999 and 2005 evaluating the effectiveness of compounds applied 2 to 21 d before harvest at reducing postharvest decay in fresh Florida citrus fruit.

Materials and Methods

Between 1999 and 2005, eleven different fungicides and a defense elicitor compound (acibenzolar-S-methyl) were tested on a total of eight different fresh citrus cultivars for their ability to provide continuing decay control after harvest. Table 1 lists the names, percent active ingredient, manufacturer, and the rate of application for the materials used. All materials were applied at the manufacturer's suggested label rate. The experimental plots were located in St. Lucie and Polk Counties within Florida. Randomized block designs with three or four replicates were used in each trial. Trials prior to 2004 were sprayed at 125 GPA (1169.2 L·ha⁻¹) using an air blast sprayer. In later studies a Stihl® (Virginia Beach, VA) backpack blower/sprayer was used to apply the materials. Sprays applied with the Stihl® backpack sprayer were applied to runoff at 3 to 4 L per tree.

Commercially mature fruit were usually harvested twice: 2–5 days (first harvest) and 2–3 weeks (second harvest) following the spray application, depending on the trial. The fruit were harvested equally from both sides of the tree between 0.5 and 2.5 m aboveground. When necessary, fruit were degreened with ~4 ppm ethylene at 85 °F (28 °C) with ~90% relative humidity.

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Table 1. Common name, trade name, percent active ingredient, manufacturing company, and rate applied for the materials tested.

Generic name	Trade name	Active ingredient (%)	Company, city, state/country	Rate applied
Acibenzolar-S-methyl	Actigard 50 WG ^z	50.0	Syngenta, Greensboro, NC	1.8 oz/acre [50 g/acre (123.6 g·ha ⁻¹)]
Azoxystrobin	Abound	22.9	Syngenta, Greensboro, NC	16 fl oz/acre (1.2 L·ha ⁻¹)
Benomyl	Benlate SP	50.0	DuPont, Wilmington, DE	2 lb/acre (2.2 kg·ha ⁻¹)
Copper hydroxide	Kocide DF	61.4	Griffin, Valdosta, GA	4 lb/acre (4.5 kg·ha ⁻¹) metallic copper
Fenbuconazole	Enable 2F	23.5	Dow AgroSciences, Indianapolis, IN	8 fl oz/acre (0.6 L·ha ⁻¹)
Fosetyl AL	Aliette WDG	80.0	Bayer CropScience, Montvale, NJ	5 lb/acre (5.6 kg·ha ⁻¹)
Phosphorous acid (fungicide)	Phostrol	53.6	AGTROL International, Houston, TX	4 pt/acre (4.7 L·ha ⁻¹)
Phosphorous acid (nutritional)	Nutri-Phite (ON-16.1P-0K) ^y	40.0	Biagro Western Sales, Inc., Visalia, CA	4 pt/acre (4.7 L·ha ⁻¹)
Pyraclostrobin	Headline	23.6	BASF, Mount Olive, NJ	16 fl oz/acre (1.2 L·ha ⁻¹)
Pyraclostrobin and Boscalid	Pristine	12.8 / 25.2	BASF, Mount Olive, NJ	18.5 fl oz/acre (1.4 L·ha ⁻¹)
Thiophanate-methyl	Topsin M4.5FL	45.0	Cerexagri, Inc., King of Prussia, PA	16 fl oz/acre (1.2 L·ha ⁻¹)
Thiophanate-methyl	Topsin M WSB	70.0	Cerexagri, Inc., King of Prussia, PA	2 lb/acre (2.2 kg·ha ⁻¹)

^zNot registered for use on citrus.

^yNot a fungicide.

The fruit were washed with an alkali surfactant solution (Fruit Cleaner 395, FMC Corp., Lakeland, FL) and waxed with a shellac wax (Sta-Fresh 590, FMC Corp.). No fungicides were applied postharvest. One trial with a new grapefruit hybrid (USDA 1-77-19) harvested after Hurricane Wilma in 2005 was not washed or waxed because the scarce fruit was also used to evaluate possible physiological disorder development.

Most fruit were stored at 50 °F (10.0 °C) with about 90% relative humidity and evaluated every 2 to 4 weeks, depending on the cultivar, for decay and the development of physiological disorders. Fruit of USDA 1-77-19 were stored at 58 °F (14.4 °C). Decay from natural infections and the development of physiological disorders were evaluated during storage. The type of decay occurring on each fruit was visually identified as either SER, anthracnose, or green mold (*Penicillium digitatum*). Other decays such as sour rot (*Geotrichum citri-aurantii*) were only rarely observed, but could be present on up to 18% of the fruit. A single fruit may exhibit more than one type of decay and/or physiological disorder. The total percentage of healthy fruit or fruit with any visible decay was also recorded. The experiments were terminated after significant decay developed. Cumulative results throughout the storage period are reported. At each evaluation, decayed fruit were removed before placing the fruit back into storage.

Data were transformed to arcsine values and analyzed by analysis of variance using SAS (PROC GLM) for PC (SAS Institute Inc., Cary, NC). When differences were significant ($P \leq 0.05$), individual treatment means were separated using Duncan's multiple range tests ($P = 0.05$). Means presented are untransformed values.

Results and Discussion

In the present studies, different compounds (mostly preharvest fungicides registered for use on citrus) were evaluated over six seasons for their ability to reduce decay on fresh Florida grapefruit, tangerines, and oranges. Not all compounds were tested every season, but were continued in our evaluations depending

on their apparent potential based on earlier tests. Phosphorous acid, copper hydroxide, fosetyl Al, and benomyl have been commercially available to citrus growers for years. Others, such as fenbuconazole, azoxystrobin, pyraclostrobin, and thiophanate methyl are relatively new. Acibenzolar-S-methyl (Actigard; Syngenta, Greensboro, NC) is reported to be an activator of plant defenses (Romero et al., 2001) that has been used for vegetables, but is not registered for use in citrus. Phosphorous acid is labeled as a fungicide (Phostrol; AGTROL International, Houston) or used as a nutritional product (Nutri-Phite; Biagro Western Sales, Inc., Visalia, CA).

All compounds were evaluated for their ability to reduce physiological disorders and decay during storage. While physiological disorders occasionally appeared on fruit in storage, their occurrence was almost never affected by the treatments (data not shown). As expected, postharvest decay was most affected by these preharvest sprays. Commercially, SER usually causes the greatest postharvest losses in fresh Florida citrus fruit (Brown and Albrigo, 1970), and during the present studies SER was most often the principal cause of decay. Therefore, the primary indicator of success for these compounds is their ability to reduce decay (usually SER) and increase the total amount of healthy fruit after storage compared to the control.

Early tests showed no significant increase in the overall numbers of healthy fruit or reductions in SER after storage with preharvest application of fenbuconazole, fosetyl-Al, phosphorous acid (Nutri-Phite), copper hydroxide, acibenzolar-S-methyl, or azoxystrobin (Tables 2 and 3). Consequently, most of these materials were excluded from further tests. Only benomyl gave significant improvement compared to the control in these early tests.

Pyraclostrobin, phosphorous acid (from a different source—Phostrol), a combination of pyraclostrobin and boscalid (Pristine), and thiophanate-methyl were subsequently tested in addition to benomyl. Of these compounds, benomyl and thiophanate-methyl most consistently increased the number of healthy fruit and reduced postharvest decay after storage, and were the fungicides tested most. Including all tangerine, orange, and grapefruit trials, benomyl and thiophanate-methyl significantly increased

Table 2. Effectiveness of different compounds applied preharvest at significantly increasing the number of healthy citrus fruit after simulated postharvest commercial storage compared to the control.

Treatment	Tangerine	Orange	Grapefruit
Benomyl	4/6 ^y	1/2	5/6
Thiophanate-methyl (WSB)	2/3	1/2	3/5
Thiophanate-methyl (FL)	1/1		
Pyraclostrobin	1/1	0/1	1/3
Phosphorus acid	1/4	0/1	0/4
Pyraclostrobin + Boscalid	0/2		
Copper hydroxide	0/3		0/1
Azoxystrobin	0/2		0/2
Fenbuconazole	0/3		0/1
Fosetyl AL	0/3		0/1
Acibenzolar-S-methyl	0/2		0/1

^xNumber of trials the treatment resulted in significantly ($P < 0.05$) more healthy fruit than the control in at least one of the two harvests.

^yTotal number of trials the material was tested.

Table 3. Effectiveness of different compounds applied preharvest at significantly reducing the incidence of stem-end rot of citrus fruit after simulated postharvest commercial storage compared to the control.

Treatment	Tangerine	Orange	Grapefruit
Benomyl	2/6 ^y	2/2	5/6
Thiophanate-methyl (WSB)	1/3	2/2	3/5
Thiophanate-methyl (FL)			1/1
Pyraclostrobin	0/1	0/1	1/3
Phosphorus acid	0/4	0/1	0/4
Pyraclostrobin + Boscalid			1/2
Copper hydroxide	0/3		0/1
Azoxystrobin	0/2		0/2
Fenbuconazole	0/3		0/1
Fosetyl AL	0/3		0/1
Acibenzolar-S-methyl	0/2		0/1

^xNumber of trials the treatment resulted in significantly ($P < 0.05$) less SER than the control in at least one of the two harvests.

^yTotal number of trials the material was tested.

the number of healthy fruit remaining after storage in 71% and 60% of the trials, respectively (Table 2), and decreased SER in 64% and 60% of the trials (Table 3). The efficacies of these two fungicides were not noticeably different among citrus cultivars. Even when there was no significant difference compared to the control, preharvest application of benomyl or thiophanate-methyl tended to result in more healthy fruit and less SER incidence after storage. Over all trials and harvest dates, preharvest application of benomyl or thiophanate-methyl resulted in a 44% and 47% reduction, respectively, of total decay after storage (data not shown). The relative efficacies of these two fungicides did not change between the first and second harvests, indicating that they reduced postharvest decay even when the harvest was delayed as much as 3 weeks (data not shown). Naqvi and Dass (1991), Ritenour et al. (2004), Smilanick et al. (2006), and Zhang and Timmer (2007) also found significant decreases in postharvest decay of citrus after preharvest application of benomyl or thiophanate-methyl. Both materials break down to form carbendazim, which is the primary source of fungicidal activity (Clemons and Sisler, 1969; Saleh and Negr, 1980).

Including all tangerine, orange, and grapefruit trials, pyraclostrobin significantly increased the number of healthy fruit after storage in a total of two out of five trials, and decreased

the SER incidence in one of the trials. However, even when not significantly different, total postharvest decay was never greater than the control and averaged 27% less than the control over all trials where pyraclostrobin was tested. While Ritenour et al. (2004) did not observe significant reductions in postharvest SER after a preharvest pyraclostrobin application, Zhang and Timmer (2007) observed on several occasions a reduction of SER incidence by pyraclostrobin, while also, pyraclostrobin was not as effective as thiophanate-methyl or benomyl. There was no significant effect of preharvest phosphorous acid on postharvest SER in any of the nine trials it was tested, but the number of healthy fruit was increased in one of the trials. The addition of boscalid with a lower concentration of pyraclostrobin (Pristine) in the preharvest treatments resulted in no significant effect on healthy fruit, and a reduction of SER incidence in one of the two trials it was tested.

Even though postharvest fruit losses were primarily a result of SER, the occurrence of anthracnose was high (>15%) after some harvests. Benomyl had been recommended as the most effective fungicide at reducing post-bloom fruit drop (PFD) in citrus (Timmer et al., 2003) caused by a different species of *Colletotrichum* (*C. acutatum*). Thiophanate-methyl has been granted a specific exemption by the U.S. Environmental Protection Agency (EPA) specifically for the control of PFD in citrus and is now recommended as the most effective fungicide for its control (Timmer et al., 2007). Wardowski and Brown (1993) reported that benomyl is only moderately effective against postharvest anthracnose. Ritenour et al. (2004) observed reductions in postharvest decay due to anthracnose after preharvest benomyl application in two tests, but not with thiophanate-methyl. However, the authors concluded that low incidence of anthracnose in their studies necessitated further tests before making definite conclusions about the efficacy of thiophanate-methyl on postharvest anthracnose. Zhang and Timmer (2007) later showed on tangerines that preharvest application of benomyl or thiophanate-methyl both significantly reduced postharvest anthracnose and SER.

Significant reductions in postharvest anthracnose were observed in the current tests on 'Ruby Red' grapefruit in 2004 (Table 4) and a new grapefruit hybrid (USDA 1-77-19) in 2005 (Table 5). Pyraclostrobin, pyraclostrobin + boscalid, thiophanate-methyl, and benomyl all significantly reduced anthracnose incidence in 'Ruby Red' grapefruit compared to the control after 8 weeks of storage (Table 4). Materials with pyraclostrobin were most effective, with pyraclostrobin and pyraclostrobin + boscalid reducing anthracnose by 84% and 71%, respectively. Thiophanate-methyl and benomyl reduced anthracnose 67% and 51%, respectively. Zhang and Timmer (2007) showed the strobilurin fungicides, azoxystrobin and pyraclostrobin, were as effective against post-

Table 4. Postharvest decay of 'Ruby Red' grapefruit after 8 weeks storage at 50 °F. The trees were sprayed with different fungicides on 20 Oct. 2004 and harvested 2 d later.

Treatment	Stem end rot (%)	Anthracnose (%)
Pyraclostrobin	13.1	6.5 d ^x
Pyraclostrobin + Boscalid	11.6	12.2 cd
Thiophanate-methyl (WSB)	4.8	13.5 cd
Benomyl	7.5	20.1 bcd
Azoxystrobin	11.9	22.6 abc
Phosphorus acid	12.8	33.6 ab
Control	6.9	41.2 a

^xValues within each column followed by unlike letters are significantly different by Duncan's multiple range test at $P < 0.05$.

Table 5. Postharvest decay of USDA 1-77-19 (grapefruit hybrid) after 4 weeks storage at 58 °F. The trees were sprayed with different fungicides on 4 Nov. 2005 and harvested 5 d later.

Treatment	Stem end rot (%)	Anthraco-nose (%)
Thiophanate-methyl (FL)	1.5 cd ^z	5.8b
Benomyl	1.7 c	15.8ab
Thiophanate-methyl (WSB)	3.9 bc	13.6ab
Pyraclostrobin	8.4 b	11.1ab
Pyraclostrobin + Boscalid	8.6 b	22.0a
Phosphorus Acid	19.3 a	13.7ab
Control	19.5 a	18.5a

^zValues within each column followed by unlike letters are significantly different by Duncan's multiple range test at $P < 0.05$.

harvest anthracnose as benomyl or thiophanate-methyl. There were no significant effects of the fungicides on SER, most likely because anthracnose was the major decay organism (Table 4).

In 2005 a new formulation of thiophanate-methyl (Topsin M 4.5FL) was tested on USDA 1-77-19, along with thiophanate-methyl, benomyl, pyraclostrobin, pyraclostrobin + boscalid, and phosphorous acid. In this case, incidence of postharvest SER and anthracnose were about equal (Table 5). All fungicides except phosphorous acid significantly reduced postharvest SER compared to the control. Thiophanate-methyl and benomyl again produced the greatest reductions (80% to 92%) in SER. Only the new formulation of thiophanate-methyl significantly reduced postharvest anthracnose, decreasing its incidence by 69%.

The above data add further evidence that preharvest application of thiophanate-methyl provides postharvest decay control similar to benomyl. Under Florida conditions, these two fungicides effectively control SER (Table 3) and may also reduce postharvest anthracnose (Tables 4 and 5).

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