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Evaluation of the Alter-Rater model for spray timing for control of Alternaria brown spot on Murcott tangor in Brazil

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Abstract

Alternaria brown spot, caused by *Alternaria alternata,* recently appeared in Brazil and is now causing serious damage to Murcott tangors in São Paulo State. The Alter-Rater model and other systems for timing fungicide applications as well as different fungicide programs were evaluated in two seasons in different citrus-growing areas of São Paulo State. Alternaria brown spot severely reduced yields of Murcotts in the unsprayed controls in both years. Fungicide applications reduced fruit drop and increased yields in many cases. Regression analyses showed that yield declined as disease severity increased. A copper fungicide applied according to the Alter-Rater model with a trigger value of 50 required one to two more applications than a calendar program in both locations in 2002–03. Use of the Alter-Rater 50 increased the number of fruit harvested in both locations, but did not improve fruit quality when compared to the calendar program. Use of the Alter-Rater with trigger values of 100 or 150, or the Copper model forecast fewer sprays and resulted in poor disease control and low yields. In a test in 2003–04, fungicide applications timed with the Alter-Rater 50 were no more effective than calendar sprays. Copper fungicides throughout the season or programs with copper applied early followed by dithiocarbamates or QoI fungicides later provided better disease control than a full-season mancozeb program. Murcott product in some citrus growing areas in Brazil will be possible only if the value of the crop is sufficient to justify 10 or more fungicide applications per year.

Keywords: *Alternaria alternata*; Disease model; Citrus

1. Introduction

Alternaria brown spot (ABS) is a serious disease of many tangerines and their hybrids in humid and semiarid citrus areas (Timmer et al., 2003). The disease produces black necrotic lesions on young leaves, twigs, and fruit. On leaves and twigs, lesions may expand rapidly due to production of a host-specific toxin by the pathogen, often resulting in leaf drop and twig dieback. On fruit, lesions vary from small dark necrotic spots to large sunken pockmarks, thereby reducing the value of the fruit for the fresh market. Fruit are susceptible to infection until about midsummer in many areas (Timmer et al., 2000b, 2005), but fruit may be susceptible for much longer in cooler climates (Garcia-Jimenez et al., 2002; Vicent et al., 2004).

The causal agent was described originally as *Alternaria citri* Ellis & Pierce (Pegg, 1966) and later designated as *A. alternata* (Fr:Fr) Keissl. pv. *citri* (Solel, 1991). Simmons (1999) described 10 new species from a world-wide collection of citrus isolates but molecular studies indicated that all small-spored isolates from citrus are similar and have been designated as *A. alternata* (Peever et al., 2004). One host-specific pathotype causes disease in tangerines and their hybrids, another pathotype is specific to rough lemon

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and Rangpur lime, whereas most small-spored isolates cause black rot of fruit regardless of whether they produce toxins (Peever et al., 2005).

ABS has expanded its worldwide distribution considerably in recent years. It has been known for many years in Australia and South Africa and was discovered in Florida in 1976 (Timmer et al., 2003). Subsequently, it appeared in Israel, Turkey, and Colombia and more recently in Spain and Italy (Bella et al., 2001). In Brazil, ABS was first found in Rio de Janeiro state (Goes et al., 2001) and subsequently became widespread in the principal citrus area in the state of São Paulo and also in Argentina (Peres et al., 2003). It has also recently been confirmed in Peru (Peres, unpublished).

The environmental conditions favorable for infection by *A. alternata* have been studied in Florida (USA). Release of conidia is triggered by rainfall and abrupt changes in relative humidity (Timmer et al., 1998). Temperatures of 23–27 °C were optimal for infection of citrus leaves and a minimum of 8–12 h of continuous leaf wetness was needed for significant infection to occur (Canihos et al., 1999). Under field conditions, rainfall significantly increased disease, but incidence was not related to the amount of rainfall (Timmer et al., 2000a). The Alter-Rater, a weather-based model for timing fungicide sprays for ABS control, was developed based on these studies. With the Alter-Rater model, a point value from 0 to 11 is assigned to each day depending on three weather factors: (i) rainfall more or less than 2.5 mm, (ii) leaf wetness more or less than 10 h, and (iii) average daily temperature below 20 °C, from 20 to 28 °C, or greater than 28 °C (Timmer et al., 2001). Point values are accumulated on a daily basis until a predefined value is reached, which triggers a fungicide application. In Florida, trigger values of 50, 100, or 150 were suggested based on the susceptibility of the cultivar, the area of the state, and the disease history of the grove.

Although some cultural measures can be used to help control ABS, fungicide applications are essential to produce high-quality fruit free of external blemishes (Timmer et al., 2000b, 2005). Of the fungicides that control ABS, copper products, mancozeb, chlorothalonil, and pyraclostrobin are registered in Brazil. Other QoI fungicides and iprodione are known to be effective and are recommended in other regions (Timmer et al., 2003, 2005). However, those products are subject to development of fungal resistance and must be alternated or mixed with protectant fungicides.

ABS is a major fruit blemish problem and is presumed to cause yield loss based on the fruit drop it causes and/or its debilitating effects on tree health. However, there is little quantitative information available on the effect of the disease or control measures on yield of citrus. In fungicide evaluations, Timmer and Zitko (1997, 1999) and Timmer (2000) found that programs controlling blemishes on fruit from ABS increased yield in one of two trials with Dancy tangerines, in one test on Minneola tangelos, and in three of four trials on Murcott tangor. However, most of these studies were conducted on young groves with 30–300 fruit per tree and may not be indicative of yield effects on mature trees.

The current study was undertaken to determine the usefulness of the Alter-Rater model for timing of fungicide sprays in the main citrus growing areas in Brazil in comparison with other fungicide programs. The effect of spray programs on yield was determined and related to the severity of disease on the fruit.

2. Materials and methods

2.1. Trials 2002–03

One trial was established in a 12-year-old planting of Murcott tangor (probable *Citrus sinensis* (L.) Osb. × *C. reticulata* Blanco hybrid) on a farm near Itapetininga in south-central São Paulo State, Brazil. Treatments evaluated were three trigger values of the Alter-Rater model, 50, 100, or 150 points, a calendar program, a program chosen by the grower, and an unsprayed control (Table 1). Kocide WDG (copper hydroxide, 35% metallic copper) at 1.0 kg $1000\,l^{-1}$ was used for all sprays. Fungicide applications were made with an airblast sprayer (Jacto, Arbus, Disc D4 and Core DC25) using $1500\,l\,ha^{-1}$ and approximately $5\,l\,tree^{-1}$. On 9 September when the spring growth flush was quarter to half expanded, applications were made to all treatments except the grower choice. Subsequently, treatments for the Alter-Rater 50, 100, and 150 trigger values were made based on the model (Timmer et al., 2001). Rainfall, leaf wetness, and average daily temperature data were obtained from an automated weather station (Adcon) located 7 km from the experimental site. Calendar sprays were applied at about 16–20 d intervals and the grower applied sprays based on historical practices. Spray dates for the treatments were: Alter-Rater 50: 9 September, 1, 18 October, 4, 25 November, 3, 17, 27 December, 6, 15 January; Alter-Rater 100: 9 September, 14 October, 25 November, 11, 27 December, 13 January; Alter-Rater 150: 9 September, 31 October, 3 December, 2 January; calendar: 9, 24 September, 10, 23 October, 8, 25 November, 13 December, 2, 15 January; grower choice: 19 September, 10 October, 14, and 25 November.

Treatments as described above were replicated three times on rows of about 50 trees each and arranged in a completely randomized design. Ten trees were selected arbitrarily in each row for evaluation in August 2003. The number of fruit on each tree was counted and 50 fruit per tree selected arbitrarily were rated for disease

Table 1
Effect of fungicide application at three different trigger values for the Alter-Rater, a calendar system, and a grower choice program on control of Alternaria brown spot of citrus in 2002–03

Location/treatment	No. of applications	Spray intervals (days)	Avg. no. of Alter-Rater points	Fruit disease severity (0–5)[a]	Marketable fruit (%)[a]	Fruit harvested (no./tree)	AUC[b] (fruit drop) 16 Dec–25 Mar
Itapetininga[c]							
Alter-Rater 50	10	7–21	55	1.4 d	61.5 a[d]	1216 a	263 b
Alter-Rater 100	6	16–42	111	3.4 b	13.9 c	467 d	295 b
Alter-Rater 150	4	30–50	160	4.1 a	5.2 cd	555 cd	257 b
Calendar	9	16–20	63	1.3 d	59.7 a	1074 b	272 b
Grower choice	4	—	—	2.7 c	34.2 b	647 c	—
Control	0	—	—	4.4 a	0.0 d	236 e	340 a
Mogi Guaçu[e]							
Alter-Rater 50	11	6–18	60	2.9 c	27.6 a	479 a	—
Copper model	5	13–31	97	4.0 ab	9.6 bc	357 ab	—
Calendar	9	13–21	68	3.5 bc	17.8 ab	327 ab	—
Control	0	—	—	5.0 a	0.0 b	23 b	—

[a]Severity rated on a scale of 0 to 5, where 0 = no lesions and 5 = severe disease. Marketable fruit are those with ratings of 0 or 1. See Materials and Methods for details.
[b]AUC = area under the curve.
[c]Treatments with Kocide WDG at 1.0 kg $1000\,l^{-1}$, $1500\,l\,ha^{-1}$.
[d]Means followed by the same letter within the same column are not significantly different according to the Waller–Duncan k-ratio t test, $P \leqslant 0.05$.
[e]Treatments with Recop through November followed by Manzate at 1.5 kg $1000\,l^{-1}$, $5000\,ha^{-1}$.

severity on the following scale: 0 = no lesions; 1 = less than 10 small lesions (<2 mm diameter) per fruit, fruit marketable as fresh; 2 = 10–30 small lesions or less than four large lesions (~5 mm diameter) per fruit; 3 = 20–40% of the fruit surface covered with lesions; 4 = 41–60% of the fruit surface covered; 5 = >60% covered. The percentage of fruit marketable as fresh, those rated 0 or 1, was calculated for each treatment. During the season, the amount of fruit drop was evaluated weekly from 16 December to 25 March. This covered the period after normal fruit drop and into autumn. Fruit that dropped were collected and counted from beneath two trees selected arbitrarily in each replicate of all treatments and the area under the curve (AUC) was calculated. The averages for each factor and the AUC for fruit drop were subjected to analysis of variance and the means separated by the Waller–Duncan k-ratio t test, $P \leqslant 0.05$. Regression analysis was used to determine the relationship between disease severity and the yield as the number of fruit per tree.

A similar experiment was conducted in a 28-year-old planting of Murcott tangor on another farm near Mogi Guaçu in southeastern São Paulo State, Brazil, about 200 km northeast of the first trial. The treatments compared were the Alter-Rater model with a trigger value of 50, a calendar program, the Copper model, and an unsprayed control (Table 1). The Copper model (Albrigo et al., 2005; Lin et al., 1999) is a system developed for timing of melanose sprays in Florida and is based on the loss of coverage by copper product due to fruit expansion and rainfall. In this test, ReCop (copper oxychloride, 50% metallic copper) at 1.5 kg $1000\,l^{-1}$ was applied to all treatments through November using $5000\,l\,ha^{-1}$. Subsequently, sprays were made with Manzate (mancozeb) at 1.5 kg $1000\,l^{-1}$ to avoid problems with phytotoxicity of copper to fruit. The spray dates for the treatments were: Alter-Rater 50: 6, 23 September, 8, 26 October, 8, 18 November, 3, 12, 19 December, 3 January, 14 January; Copper model: 9, 20 September, 8 October, 1 November, 3 December; calendar: 6, 20 September, 8, 24 October, 8, 23 November, 6, 20 December, and 9 January. Data for timing of the Alter-Rater sprays were collected manually using a rain gauge, a maximum–minimum thermometer, and visual observation of dew and moisture resulting from rainfall.

Plot design was the same as the test at Itapetininga, except that only two replications were used. Fruit harvest was in July 2003 and the evaluation and statistical analyses were conducted as in the previous test.

2.2. 2003–04 trial

In 2003–04, a trial was conducted again in the Murcott planting near Mogi-Guaçu in southeastern São Paulo State. Five spray programs were applied based on the Alter-Rater 50 timing and two programs were applied based on a calendar schedule. Sprays based on the Alter-Rater 50 were applied on 29 September, 13, 31 October, 14, 25 November, 6, 24 December, 13 and 29 January. Those based on the calendar were applied on 29 September, 13, 29 October, 12 November, 3, 22 December, 5 and 29 January. Most of the programs used copper fungicides early in the year and other products later when the weather was hotter (Table 2). The intent was to find programs and timings that maximized control of ABS and minimized copper damage that occurs primarily in hot weather. In one treatment, mancozeb alone was applied to avoid any damage by copper fungicides. Copper damage was rated on a scale of: 0 = no lesions; 1 = less than 10 lesions per fruit; 2 = 10–30 lesions per fruit; 3 = 20–40% of the surface area covered; 4 = 41–60% of the surface area covered; 5 = >60% of the surface area covered.

The experimental design, the application equipment, and the method of collecting weather data and statistical analyses were the same as those used in this location the previous year. Likewise, harvest, fruit evaluation, and data analysis were conducted as in the previous year. Copper phytotoxicity was evaluated on the same scale as that used for ABS and analyzed similarly.

3. Results

3.1. 2002–03 trials

Applications were generally made on a timely basis and the number of Alter-Rater points between applications approximated those planned in both Itapetininga and Mogi Guaçu (Table 1). The number of days between the Alter-Rater sprays varied much more than the number between calendar sprays as would be expected. The number of sprays applied by the grower corresponded with the number forecast by the Alter-Rater 150 in Itapetininga. The number of sprays applied with a 2- to 3-week calendar spray was similar to that forecast by the Alter-Rater 50.

Disease was extremely severe in the grove near Itapetininga in 2002–03, resulting in no marketable fruit in the unsprayed control. Control was inadequate with severe disease on fruit and a low percentage of marketable fruit where 4–6 sprays were applied in the grower choice, Alter-Rater 100 or 150. However, the grower choice program with 4 sprays provided better control than the Alter-Rater 100 or 150 with 4 or 6 sprays. The calendar program with 9 sprays was as effective in reducing disease severity and increasing the percentage of marketable fruit as the Alter-Rater 50 with 10 sprays.

Treatment effects on yield were similar to their effects on fruit quality (Table 1). However, with the Alter-Rater 50, yields were significantly greater than those with the calendar program. Yields of trees treated with

Table 2
Effect of different spray programs on control of Alternaria brown spot of citrus in 2003–04

Timing	Program[a]									Disease severity (0–5)[b]	Marketable fruit (%)[b]	Phytotoxicity rating (0–5)[b]	Yield (fruit/tree)[b]
1. Alter-Rater	K	K	K	K	K	K	K	K	K	0.70 b	83 a	0.75 a	884 bc
2. Alter-Rater	Cop2	Cop2	Cop2	Cop2	Cop2	Cop2	Cop2	Cop2	Cop2	0.55 b	85 a	0.8 a	1762 a
3. Alter-Rater	Mz	Mz	Mz	Mz	Mz	Mz	Mz	Mz	Mz	2.75 a	32 b	0 b	1008 abc
4. Alter-Rater	Cop1.5	Cop1.5	Cop1	Cop0.75	Cop0.75	Com	Mz	Com	Mz	0.6 b	88 a	0.05 b	1386 ab
5. Alter-Rater	Cop1.5	Cop1.5	Cop 1	Cop0.75	Cop0.75	Com	Sc	Com	Sc	0.85 b	78 a	0.55 ab	1198 abc
6. Calendar	Cop1.5	Cop1.5	Cop1.5	Cop1.5	Cop1.5	Cop1.5	Cop1.5			0.8 b	84 a	0.55 ab	1086 abc
7. Calendar	Cop1.5	Cop1.5	Cop1.5	Cop1.5	Cop1.5	Mz+Com	Mz+Com			1.2 b	70 a	0.4 ab	984 abc
8. Control	No spray									1.65 ab	61 ab	0.0 b	401 c

Means followed by the same letter within the same column are not significantly different according to the Waller–Duncan k-ratio t test, $P \leqslant 0.05$.

[a]K = Kocide (copper hydroxide) at 1.0 kg 1000 l^{-1}; Mz = Manzate or Dithane (mancozeb) at 1.5 kg 1000 l^{-1}; Com = Comet (pyraclostrobin) at 0.15 kg 1000 l^{-1}; Sc = Score (difenoconazole) at 0.21 1000 l^{-1}; Cop = copper oxychloride as Cobox at 0.75, 1.0, 1.5, or 2.0 kg 1000 l^{-1}.

[b]Severity rated on a scale of 0–5, where 0 = no lesions and 5 = severe disease. Marketable fruit are those with ratings of 0 or 1. See Materials and Methods for details.

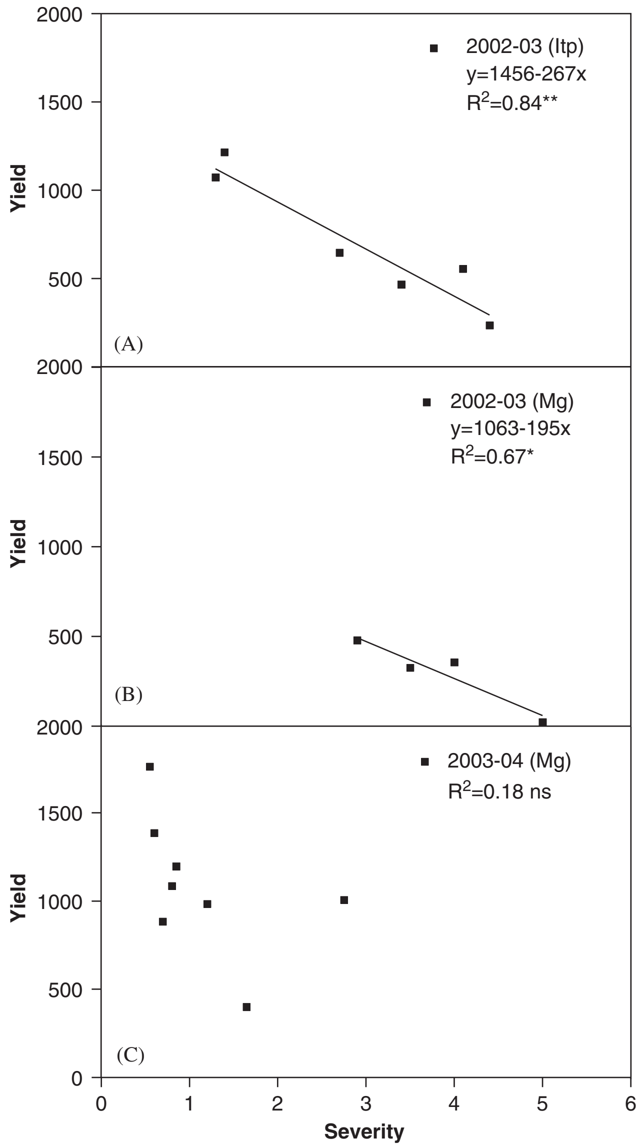

Fig. 1. Relationship between the severity rating of Alternaria brown spot and the yield as number of fruit per tree in the fungicide trials: (A) Itapetininga, 2002–03, (B) Mogi-Guaçu, 2002–03, (C) Mogi-Guaçu, 2003–04. **, *, or ns are regression coefficients significant at $P \leqslant 0.01$, $\leqslant 0.05$ or not significant, respectively.

the Alter-Rater 100 with 6 sprays were no better than with the Alter-Rater 150 with 4 sprays. There was a strong negative relationship between disease severity on the fruit and yield recorded as the number of fruit per tree ($R^2 = 0.84$, $P \leqslant 0.0001$) (Fig. 1A). Peaks in fruit drop occurred in early January and in February (data not shown). Virtually no fruit drop occurred after March in any of the treatments. Fruit drop was greatest in the non-sprayed control and significantly less in treatments that had fungicide sprays, but there were no differences among spray treatments (Table 1).

In the 2002–03 trial in Mogi Guaçu, the number of sprays applied using the Alter-Rater 50 was about the same as with the calendar program (Table 1). However, the spray intervals were more variable with the Alter-Rater 50, as expected. Disease was very severe at this location and all of the remaining fruit on the unsprayed control had a rating of 5.0. The five sprays applied according to the Copper model did not significantly decrease severity ratings or increase the percentage of marketable fruit compared to the unsprayed control. The calendar and the Alter-Rater 50 programs reduced disease severity compared to the control, but only the Alter-Rater 50 resulted in a significant increase in the percentage of marketable fruit. The 11 sprays applied according to the Alter-Rater 50 resulted in only 27% of the fruit useable for the fresh market.

At Mogi Guaçu, only the Alter-Rater 50 significantly increased fruit yield compared to the unsprayed control (Table 1). However, yield with the Alter-Rater 50 was not significantly different from other treatments. There was also a negative relationship between disease severity and yield in this location ($R^2 = 0.67$, $P = 0.023$) (Fig. 1B).

3.2. 2003–04 trial

Disease was less severe in 2003–04 than it was in the previous year. Nevertheless, none of the treatments reduced disease severity on fruit or increased the percentage of marketable fruit in comparison to the unsprayed control (Table 2). However, the abscission of severely infected fruit in the control before harvest may have resulted in lower severity ratings in the control. Disease severity was greater on trees sprayed with mancozeb than on trees treated with copper fungicides early in the season and non-sprayed control. Substituting mancozeb or other non-copper fungicides for copper fungicides late in the season did not affect disease severity or the percentage of marketable fruit in comparison to full-season sprays of copper fungicides. There were no significant differences between spray programs applied on a calendar basis and those timed using the Alter-Rater model.

Stippling due to copper phytotoxicity occurred in all treatments that included copper fungicides (Table 2). There were no differences in phytotoxicity between programs that used copper fungicides throughout the season and those where other fungicides were substituted late in the season.

Yield was very low in 2003–04 in the unsprayed control (Table 2). Only two treatments significantly increased the yield compared to the unsprayed control—a full-season program of copper fungicides and a copper–mancozeb–pyraclostrobin program applied on the Alter-Rater 50 schedule.

Yields tended to be lower in treatments with higher disease severities, but the relationship was not significant when all treatments were included ($R^2 = 0.18$, $P = 0.11$)

(Fig. 1C). However, if the mancozeb treatment was removed from the regression, there was a significant negative relationship between disease severity and yield ($R^2 = 0.42$, $P = 0.01$) (data not shown).

4. Discussion

In Florida studies, use of the Alter-Rater often resulted in fewer sprays or in better disease control than use of a calendar program or the Copper model (Bhatia and Timmer, 2003). In São Paulo, the use of the Alter-Rater required one or two more sprays than a calendar program without an increase in the percentage of marketable fruit. However, yield was often higher in plots sprayed with the Alter-Rater model. The Alter-Rater 50 trigger value is essential in those citrus growing areas in São Paulo State because disease is so severe that Alter-Rater 100 or 150 trigger values resulted in far too few sprays for adequate control. As in Florida (Bhatia and Timmer, 2003), the Copper model forecast too few sprays for effective control of ABS. The Alter-Rater appears to be effective in Brazil, but a lower trigger value would have to be used to produce a high yield of marketable fruit.

Choice of fungicides is important in achieving good control of ABS. Copper fungicides are highly effective for control of this disease and the Alter-Rater model was validated using these products (Bhatia and Timmer, 2003; Timmer and Zitko, 1997, 1999). Iprodione effectively controlled ABS in Israel (Solel et al., 1997) and South Africa (Schutte et al., 1992), but resistance developed in Israel and its use is limited to one or two applications per year (Solel et al., 1996). Tebuconazole was effective in South African studies (Schutte et al., 1992), but this fungicide and other triazoles were not effective in studies in Florida (Timmer and Zitko, 1997), nor in Israel (Solel et al., 1997). Mancozeb provided good control of ABS in South Africa (Schutte et al., 1992). QoI fungicides are highly effective for ABS control and recommended in Florida, but are subject to development of resistance (Timmer and Zitko, 1997; Timmer et al., 2005). Although copper fungicides are effective and economical, they can produce stippling damage, especially if applied in hot, dry weather or in complex tank mixes with other products (Albrigo et al., 1997). Thus, other fungicides need to be substituted at some times during the season. We were unsuccessful in avoiding phytotoxicity on fruit by substituting other fungicides for copper late in the season in the 2003–04 test. Consequently, copper products may only be used early in the season for control of ABS in some citrus areas in Brazil. A full-season program with mancozeb, however, was not highly effective for disease control in our test. Mancozeb apparently has too short a residual effect or is not sufficiently active against *A. alternata* to provide a high degree of control unless applied frequently. It appears that a program using copper fungicides early and alternating pyraclostrobin, which is now registered for citrus in Brazil, and mancozeb may be the most effective program.

It is clear that failure to control ABS results not only in heavily blemished fruit but also in reduced yield due to fruit drop throughout the spring and much of the summer. In previous studies on Murcotts, Dancy tangerines, and Minneola tangelo in Florida (Timmer and Zitko, 1997, 1998; Timmer, 2000), fungicide applications increased yields of tangerine and their hybrids as well as improved fruit quality. We analyzed data published by Timmer (2000) and Timmer and Zitko (1997, 1999) and found a significant negative relationship between disease severity and yield in the Murcott trials in 1995, 1996, and 1998, in the 1991 trial with Dancy tangerine, and in the 1994 test with Minneolas, but not in the 1997 test with Murcotts nor in the 1993 test with Minneolas (data not shown). All of the tests in Florida were conducted on young trees. In the tests in Brazil, failure to spray resulted in a nearly complete loss of yield at the Mogi-Guaçu location in 2002–03 and low yields in the other trials. In both tests in 2002–03, there was a highly significant negative linear relationship between disease severity and yield. In 2003–04, the regression coefficient was much lower if all treatments are included. However, if the mancozeb only program was eliminated from the analysis, the regression coefficient was much higher. There was also a positive linear relationship between the number of applications of fungicide and yield: Itapetininga 2002–03 ($R^2 = 0.80$, $P = 0.005$); Mogi Guaçu 2002–03 ($R^2 = 0.84$, $P = 0.08$). At Mogi Guaçu in 2003–04, a large number of applications were used in all treatments and regression analysis was not possible. Thus, it is clear that fungicide applications yield on mature trees in Brazil as it does on young trees in Florida due primarily to a reduction in the abscission of infected fruit. Furthermore, we suspect that continuous defoliation and twig death caused by ABS spot may also impact on yield.

ABS appeared in Brazil very recently (Goes et al., 2001; Peres et al., 2003) and has become a devastating problem on susceptible cultivars. Very intense spray programs are obviously needed to produce fruit for the fresh market. The value of the fruit in many years is insufficient to support the cost of such programs. Murcott is usually grown for the fresh market and where the value warrants, sprays timed using the Alter-Rater model can produce satisfactory results. Murcott is also grown for juice in Brazil and is added to orange juice to improve the color. For this use, blemishes are unimportant. Low rates of inexpensive copper fungicides could be applied to reduce yield loss using the Alter-Rater model for timing. Nevertheless, many growers in Brazil are replacing groves of susceptible cultivars with resistant ones.

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