

After-infection Activity of Protective Fungicides against Apple White Rot

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In a trial to select suitable fungicides for developing a spray program that can control apple white rot effectively, after-infection activities in some protective fungicides were detected. Six fungicides, mancozeb, propineb, benomyl, folpet, azoxystrobin and iminoctadine-triacetate, which had been extensively used in apple orchards, were sprayed on 12-year-old apple trees (cv. Fuji) at 15-day intervals from late May to late July. Disease incidences and infection frequencies of the fruit bagged just before and soon after each spray were examined. When the infection frequency or disease incidence of the fruit bagged after each spraying of fungicide was significantly lower than those of the fruit bagged before spraying, the fungicides appeared to confer after-infection activity. The six fungicides showed diverse activities on white rot: folpet showed after-infection activity on disease development, iminoctadine-triacetate showed after-infection activity on infection, azoxystrobin showed after-infection activity on disease development and infection, and mancozeb, propineb and benomyl showed no distinct activity. The activity of a fungicide became much higher when it was sprayed alternately with other fungicide rather than successive spraying of the same fungicide. Analysis of the properties of these protective fungicides could lead to the development of a highly effective spray program against white rot.

Keywords : apple white rot, after-infection activity, protective fungicides

Apple white rot, a disease caused by *Botryosphaeria dothidea* (Moug.) Ces. & De Not, is one of the most serious apple disease in Korea (Uhm, 1998; Lee et al., 2006) where the highly susceptible cv. Fuji accounts for more than 70% of apples produced. In 1998, as an extreme example, almost 50% of Fuji apples decayed before harvest because of this disease, and some farmers even gave up the harvest (Lee et al., 2006). The major inoculum source of this disease is the pycnidiospores that are produced in warts formed on stems

infected with the causal fungus (Fulkerson, 1960; Hayashi, 1984; Kim et al., 1995; Ogata, 1997; Sutton, 1981). The spores are dispersed whenever precipitation reaches more than 2 mm for more than 2 h during the apple-growing season (Ogata, 1997). Chung et al. (1993) reported that 6 to 360×10³ spores were found on the surface of an apple (cv. Fuji) picked about 10 h after rainfall in mid-July. Because apple fruits grow in close proximity to the inoculum source, they can easily be infected by the fungus, causing white rot. The infection can occur continuously from two weeks after petal fall to late August, yet the symptom do not appear until six or eight weeks before harvest (Drake, 1971; Kohn and Hendrix, 1983) at which time fungicidal sprays are usually terminated. This fact that symptom develops after termination of chemical spray makes it difficult to assess the control efficacy of each fungicide sprayed during the possible infection period.

Several control measures, such as chopping prunings left on the orchard floor, elimination of warts on the stems, and coating of the diseased branch or trunks with polymer to prevent the spores from being dispersed, have been proposed (Sutton, 1990). Although these control measures based on cultural practices are not effective enough to control the diseases, they can be used to supplement the efficacy of fungicides.

In Korea, most apple growers spray protective fungicides at 10-day intervals from mid-June, and some growers shorten the spray interval to less than 10 days in the rainy season (Uhm, 1998). Despite this frequent spraying, considerable economic damage from white rot is inevitable each year.

In this study, it was tried to develop a spray program that could reduce the disease incidence with reduced frequency of chemical spraying. For reducing spray frequency, extension of spray intervals or early termination of chemical spraying is required. In the conventional spray program, fungicidal spraying is recommended to continue until mid- or late-September for late-season varieties (Uhm, 1998). For the first step in developing the spray program, selection of fungicides applicable to a 15-day-interval spray program was conducted. Because the control efficacy of the fungicides against white rot might be fluctuated in accordance

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with the apple-growing season, trials to select the most suitable chemicals for each growth stage of the apples were conducted. During the course of this experiment, after-infection activities of the protective fungicides against white rot were detected. A series of experiments were conducted to confirm the after-infection activities of several fungicides that might open a new approach for controlling the disease.

Materials and Methods

Fungicides used. In trials conducted in 1997, six fungicides were selected, all of which have been used intensively to control white rot: mancozeb (Dithane M-45 75WP; Kyung Nong Co., Seoul, Korea), propineb (Antracol 70WP; Dongbu Hannong Chemical, Seoul, Korea), benomyl (Benlate 50WP; Dongyang Chemical, Seoul, Korea), folpet (Folpet 50WP; Hankook Samkong Co., Seoul, Korea), iminoctadine-triacetate (Befuran 25LC; Misung Ltd., Pyungtaek, Korea) and azoxystrobin (Amistar 10WP; Dongbu Hannong Chemical, Seoul, Korea).

Experimental orchard and chemical application. The experiments were conducted in a commercial orchard using 12-year-old cv. Fuji apple trees on M.26 rootstock near Daegu City. Almost all trees were severely infected with *B. dothidea* and had numerous warts on their branches and even on small twigs. The fungicides were applied at the manufacturer's recommended dosage of each chemical for the control of apple white rot, 1.5, 0.325, 1.4, 1.0, 0.1 and 0.25 g. a.i/L for mancozeb, benomyl, propineb, folpet, azoxystrobin and iminoctadine-triacetate, respectively. These dosages were applied with a single-nozzle spray gun at 3.5 MPa to runoff. A basic spray was applied in the experimental orchard comprising iminoctadine-triacetate before bloom and Systhane M, a combined formula of mancozeb and myclobutanil, at petal fall.

Examination of disease incidence and latent infection. The diseased fruit were counted at weekly intervals from late August to early October. Because the apples were still in bags, the diseased fruit were counted among those that had been shed. After removing the fruit bags in early October, diseased fruit were examined at five-day intervals until harvest around late October/early November. At harvest, symptomless fruit without visible mechanical or insect wounds were placed on a polystyrene egg carton in a cardboard box and incubated at 25°C. Disease incidence was determined weekly for four weeks. Because most of the infected fruit developed symptoms during this period, the recorded disease incidence was assumed to reflect latent infections.

Assessment of after-infection activities of fungicides against white rot by successive application of the same fungicide. Chemical spraying was initiated on June 12 and was repeated four times at basically 15-day intervals. To detect after-infection activity, 100 fruits were bagged just before and soon after each spray. The fruit bags (Nonghyup Agro Inc., Daegu, Korea) were two-layered with a paraffined waterproof inner layer that protected the fruit from additional infection by natural inoculum and from exposure to additional chemical spraying. The bagging of fruit enabled control efficacies of the fungicides to be assessed by one application. To detect infections that occurred in between each spraying time, 100 fruits on fungicide-untreated trees were bagged at the time of each spraying.

Assessment of after-infection activities of fungicides against white rot by alternated application with other fungicide. Although the time of infection in apple white rot has been controversial (Drake, 1971; Kohn and Hendrix, 1983; Parker and Sutton, 1993b), Kim and Uhm (2004) confirmed that 11.0% of the fruit were infected until late May. Therefore, the application of chemicals to control white rot was determined to begin before late May. On the basis of this concept, fungicide-spraying programs of 15-day intervals beginning in late May or early June and continuing until late July were prepared with several fungicides, including the three fungicides, azoxystrobin, iminoctadine-triacetate and folpet, which had shown after-infection activity in the previous trial conducted by successive spraying of the same fungicide. However, because four to five fungicides were required to cover the possible infection period of white rot, we also used propineb, which had also shown post-infection activity in the early growing season (Table 1).

Manufacturer's recommended dosage of captan (Captan 50WP; Kyung Nong Co., Seoul Korea) and Systhane M, a combined formula of myclobutanil and mancozeb (Kyung Nong Co., Seoul Korea), were sprayed before blooming and at petal fall, respectively (Table 1). Because bagging was necessary for the detection of post-infection activity, application of the test chemicals was initiated when the fruit were big enough for bagging. In the 1998 trial, the fruit were too small to bag at the time of the third cover spray in late May, and application of propineb was postponed until the fourth cover. Instead, thiram (Sulmane 80WP; Hankook Samkong Co., Seoul Korea) was sprayed at the third cover for possible control of *Alternaria blotch* and *Marssonina blotch* (Table 1). In the trial in 1999, the fruit size reached bagging stage in late May and application of the test chemical was initiated from the third cover, and the spray sequence was slightly changed to enhance the control effectiveness against *Marssonina blotch*. At the seventh

Table 1. Spray programs for detecting after-infection activities of the fungicides by alternate application with other fungicides

Ser. No.	1998 y		1999	
	Date Sprayed	Fungicides	Date Sprayed	Fungicides
1	15 Apr	Captan	18 Apr	Captan
2	6 May	Systhane M	13 May	Systhane M
3	22 May	Thiram	28 May	Propineb
4	7 Jun	Propineb	12 Jun	Azoxystrobin
5	21 Jun	Iminoctadine-triacetate	26 Jun	Iminoctadine-triacetate
6	6 Jul	Folpet	11 Jul	Folpet
7	22 Jul	Azoxystrobin	26 Jul	Iminoctadine-triacetate

cover spray on July 26, iminoctadine-triacetate was sprayed again to control *Marssonina* blotch; however, bagging was not performed. These spray programs were applied to 10 trees in the experimental orchard described above. To detect the post-infection activity of each fungicide, 100 fruits from 10 trees were also bagged before and after spraying of each test chemical. When bagging fruits, attention was paid to make sure the bagged fruit was uniformly distributed over the entire tree.

Design of experiments and analyses of data. All of the experiments in this study were conducted in a randomized complete block design with single tree replicates. Data were analyzed with a *t*-test to detect differences in disease incidences and infection frequencies between the fruit bagged before and after each spray. If the infection frequencies or disease incidences of the fruit bagged after spraying were significantly lower than the fruit bagged before spraying, it was determined that the fungicide conferred after-infection activity.

Results and Discussion

Disease incidence and infection frequency in fungicide-

untreated plots. Disease incidence and infection frequency of white rot in fruit that was periodically bagged at 15-day intervals in a fungicide-untreated plot over three successive years from 1997 are shown in Figure 1. In 1997, 32.1% of the fruits had already been infected by June 12, when the experiment was initiated. Of these, 11.1% were diseased by harvest and the remaining 21% were latently infected (Fig. 1). The timing of infection for this disease is controversial because Kohn and Hendrix (1983) asserted that the time of infection is four to six weeks before harvest, whereas Parker and Sutton (1993b) believed it to be seven weeks after petal fall, and Drake (1971) reported that infection may occur soon after fruit set. In this study, at least 32.1% of the apples were already infected before June 12, about four weeks after petal fall, supporting the concept that infection may occur soon after fruit set (Drake, 1971). The infection frequency increased consistently, reaching 60.8% on June 28, 98.4% on July 14, with all of the fruit being infected by July 29 (Fig. 1).

The infection frequency increased by 28.7% between June 12 and June 28; however, disease incidence increased by 19.3% during the same period (Fig. 1). The difference between infection frequency and disease incidence was greater thereafter. During the 16 days from June 28 to July

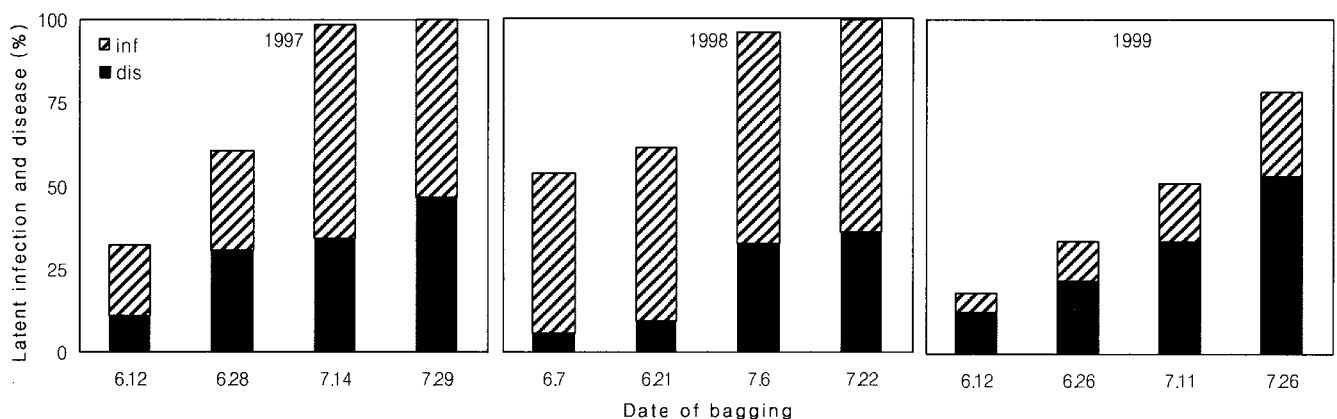


Fig. 1. Incidence of latent infection and disease incidence of white rot in fungicide untreated plot determined by bagging of the fruit periodically (1997-1999).

14, infection frequency increased by 37.6% and disease incidence increased by only 4% (Fig. 1). During the two weeks from July 14 to July 28, infection frequency increased by 1.4%, whereas disease incidence increased by 12.2% (Fig. 1). These differences in the increase rate of infection frequency and disease incidence could be explained by multiple infections of the fruit because the frequency of infection in this experiment was cumulative until bagging. The infection frequency of the bagged fruit on July 14 was 98.4%, which allowed almost no room for an increase in infection frequency in the 14 days from July 14 to July 28; however, disease incidence increased by 12.2% during this period (Fig. 1). Therefore, the increase in disease incidence was probably caused by multiple infections on some fruit.

In 1998, 53.6% of apples were infected until June 6 when bagging was initiated and, among these, 5.9% were diseased until harvest (Fig. 1). The infection frequency had increased to 99.5% by July 29 and 36.5% of these fruit were diseased until harvest (Fig. 1). In 1999, the infection frequency of 18.8% detected by the first bagging conducted on June 12 was far lower than that of the two previous years; however, the proportion of disease incidence to infection frequency was higher than the two previous years, revealing that about 66.0% of fruits were diseased (Fig. 1). The cumulative infection frequency until the last bagging on July 26 was 78.2%, of which 52.9% were diseased until harvest (Fig. 1). Great variation in the ratio of infection frequency and disease incidence for each year was observed, the cause of which may be very difficult to determine.

Postinfection activities of fungicides against apple white rot determined by successive spraying of the same fungicide. The infection frequencies of the fruit bagged before chemical treatment on June 12, which was the initiation date of this experiment in 1997, were roughly similar to those of the fruit bagged at same time in the untreated plot (Fig. 1). However, the disease incidence in the fungicide-untreated plot was much higher than that of the treated plots (Table 2). This might be caused by a lowering of metabolic activity because of heavy defoliation by Marssonina blotch in the untreated plot. The six fungicides showed diverse activities on white rot. However, at the beginning of this trial on June 12 when the infection frequencies were still low, all of the fungicides except mancozeb showed after-infection activity (Table 2).

Mancozeb, which has long been used for the control of white rot and other major apple diseases in Korea, showed low after-infection activity, with the infection frequencies not significantly reducing in all treatments. Moreover, mancozeb had very low protective efficacy against white rot when it was sprayed at 15-day intervals. In particular, during the 16 days from June 28 to July 14 when the infection frequency rapidly increased in the untreated plot (Fig. 1), the infection frequency of the fruit treated with mancozeb increased from 37.9% to 89% (Table 2). Because of low after-infection activity, the infection frequency of the fruit bagged after spraying on July 29 was as high as 81.3% (Table 2). Additionally, disease incidence was not significantly reduced by mancozeb, except for one application on July 29, after which disease incidence reduced from 35.8%

Table 2. Infection frequency and disease incidence of apple white rot in each fungicide-applied plot at indicated investigation

Date Sprayed	Time Bagging	Mancozeb		Benomyl		Propineb		Folpet		Azoxystrobin		ITA ^a	
		Inf.% ^b	Dis.% ^c	Inf.%	Dis.%	Inf.%	Dis.%	Inf.%	Dis.%	Inf.%	Dis.%	Inf.%	Dis.%
12, Jun	before ^d	26.0	3.2	29.3	4.3	22.3	4.1	34.3	4.2	21.5	5.2	28.6	3.3
	after ^e	22.6	2.2	17.4	3.1	12.1	1.0	13.6	1.5	13.1	1.3	7.6	1.3
	t-test	ns	Ns	*	Ns	**	*	**	*	**	*	**	ns
28, Jun	before	49.5	10.1	45.0	7.0	51.8	17.6	36.2	5.3	39.2	2.5	43.8	11.2
	after	37.9	8.4	42.5	6.9	48.9	10.4	29.6	2.2	27.1	1.2	13.5	2.1
	t-test	ns	Ns	Ns	Ns	Ns	ns	ns	**	**	*	**	**
14, Jul	before	89.0	21.0	81.9	16.7	100	16.1	45.4	11.1	71.2	10.1	67.0	8.2
	after	80.3	20.3	74.2	12.0	55.3	10.5	37.1	3.4	47.4	6.3	32.0	8.0
	t-test	ns	Ns	Ns	Ns	**	ns	ns	**	**	**	**	ns
29, Jul	before	90.3	35.8	83.1	16.8	73.1	18.3	54.9	15.9	77.7	13.4	84.8	14.6
	after	81.3	23.1	72.9	24.3	64.3	13.6	48.1	11.1	38.3	13.1	53.4	10.4
	t-test	ns	**	Ns	*	Ns	ns	ns	*	**	ns	**	ns

^aEach fruit was bagged at a indicated day before or after fungicide application.

^bIminoctadine-triacetate was abbreviated to ITA.

^cFigure indicated as infection frequency.

^dFigure indicated as disease incidence.

to 23.1% (Table 2).

Benomyl has also long been used mainly for the control of white rot, and a high after-infection activity was expected, as it is a systemic fungicide. However, it showed poor after-infection activity and no significant reduction in infection frequency was detected by applications after June 12 when large-scale infection occurred (Table 2). The protective efficacy of benomyl was also very poor because the infection frequency increased from 42.5% to 81.9% over the 16 days from June 28 to July 14 (Table 2). Because of the low after-infection activity of benomyl, the infection frequency continuously increased to reach 72.9% in the fruit bagged after the final spray on July 29 (Table 2). Disease incidence was also not significantly reduced by treatment with benomyl and even increased significantly after the treatment on July 29 (Table 2). The increase in disease incidence after treatment with benomyl was presumed to be caused by activation of latent infection.

Propineb has also been extensively used to control white rot and showed low after-infection activity in this experiment. Infection frequency was not significantly reduced by treatment with propineb except on June 12 and July 14 (Table 2); however, the former application was conducted before large-scale infection had taken place. The protective efficacy of propineb was also poor because the infection frequency of the fruit bagged just before each spray was usually much higher than that of the fruit bagged after each previous spray. In particular, after the treatment on June 28, infections drastically increased to reach complete infection of the bagged fruit just before spraying on July 14 (Table 2). Although propineb did not reduce disease incidence except for the treatment on June 12, disease incidence was not greatly increased despite the rapid increase of infection. The disease incidence of the fruit bagged after the last spray conducted on July 29 was 13.6% (Table 2), which was much lower than the fruit treated with mancozeb and benomyl.

Folpet also did not reduce the infection frequencies except for the first treatment on June 12; however, the increased rate of infection during each interval between each spray was lower than that of the other chemicals used in this experiment. The low increase in rate of infection was derived from the high protective activity of this chemical. In addition to having a high protective activity, folpet significantly reduced disease incidence in each treatment (Table 2). However, it is difficult to explain how folpet can reduce disease incidence despite its inability to reduce infection frequency.

Azoxystrobin showed high after-infection activity and poor protective activity. Infection frequencies increased at high rates during the intervals between each spray; however, they were significantly reduced by each treatment

(Table 2). Therefore, increases and decreases in infection frequencies were repeated by each treatment. In addition to high after-infection activity on infection, azoxystrobin also significantly reduced disease incidence except for the treatment on July 29 (Table 2).

Iminoctadine-triacetate also showed poor protective activity; however, it showed a high after-infection activity. The infection frequencies increased during the intervals between each spray; however, these were significantly reduced by the next treatment (Table 2). Therefore, increases and decreases in infection frequencies were repeated as was seen for treatment with azoxystrobin. Iminoctadine-triacetate, however, did not reduce the disease incidence except for the treatment on June 28 (Table 2).

From the results of the above trial, it was elucidated that the three chemicals, folpet, azoxystrobin and iminoctadine-triacetate, suppressed infection and/or symptom development in the infected fruit. Because the reductions in infection frequency and disease incidence occurred in already-infected fruit, the activities shown by the three chemicals can be regarded as after-infection activities. It was supposed that the properties of the fungicides may greatly alter the ideas about control of this disease. By actually using them in a spray program, we examined the ability of these three fungicides to suppress infection and disease development in apple white rot.

Assessment of after-infection activities of fungicides against apple white rot by alternate application with other fungicides. In 1998, 53.6% of apples in the fungicide-untreated plot were infected with white rot by June 7 (Fig. 1), whereas in the fungicide-treated plot, 17.9% of apples were infected by June 7 (Table 3). This might be caused by the application of thiram to the fungicide-treated plot at the third cover on May 22. The initial infection frequency of 17.9% in the fungicide-treated plot was reduced to 10.4% by the application of propineb on June 7; however, frequency increased to 58.4% over the following 14 days. A highly significant reduction from 58.4% to 35.2% was obtained by the application of iminoctadine-triacetate on June 21. The infection frequency was drastically increased again to 83.7% over the following 15 days (Table 3). These increases in infection frequency were presumed to be caused by the frequent rainfall for nine of the 15 days, with 170 mm precipitation from June 21 to July 6 (Korean Meteorological Administration, 1998), and the low protective efficacy of the iminoctadine-triacetate, which had already been observed in the trials in 1997 (Table 2). The application of folpet on July 6 also significantly reduced the infection frequency; however, the rate of reduction was relatively low compared with the other two fungicides. The low curative efficacy of folpet was also

Table 3. Infection frequency and disease incidence of apple white rot at indicated investigation in each plot applied fungicides according to spray program

1998					1999				
Date Sprayed	Fungicides	Time bagging	Inf. % ^a	Dis. % ^b	Date Sprayed	Fungi-cides	Time bagging	Inf. %	Dis. %
7 Jun	Propineb	before ^c	17.9	4.2	28 May	Propineb	before	9.7	1.2
		after ^d	10.4	2.2			after	2.5	0.0
		t-test	*	*			t-test	*	**
21 Jun	ITA ^c	before	58.4	27.4	12 Jun	Azoxystrobin	before	2.3	0.0
		after	35.2	2.3			after	0.0	0.0
		t-test	**	**			t-test	**	**
6 Jul	Folpet	before	83.7	26.1	26 Jun	ITA	before	35.2	8.8
		after	61.6	8.1			after	10.5	0.0
		t-test	**	*			t-test	*	**
22 Jul	Azoxystrobin	before	67.7	10.2	11 Jul	Folpet	before	23.5	7.5
		after	18.2	0.0			after	8.1	2.5
		t-test	**	**			t-test	**	**

^aEach fruit was bagged at a indicated day before or after fungicide application according to each spray program in 1998 and 1999.

^bIminoctadine-triacetate was abbreviated to ITA.

^cFigure indicated as infection frequency.

^dFigure indicated as disease incidence.

observed in the 1997 trial (Table 2). However, because of the high protective efficacy of folpet, which had also been confirmed in the previous trial, the infection frequency increased slightly from 61.6% to 67.7% over the 15 days after it was sprayed, despite frequent rainfall for 11 days with 95 mm precipitation (Korean Meteorological Administration, 1998). The infection frequency was drastically reduced again from 67.7% to 18.3% with the application of azoxystrobin on July 22 (Table 3).

Disease incidence also increased during the intervals between sprays; however, it was greatly reduced by each fungicide application. Azoxystrobin completely suppressed disease incidence. Iminoctadine-triacetate, which had shown a poor ability to reduce disease incidence in the trial with successive spraying in 1997, also greatly reduced disease incidence from 27.4% to 2.3% (Table 3), when it was used alternately with other fungicides.

In 1999, the accumulated infection frequency by late July in the untreated plot was 74.7%, which was much lower than the previous two years in which almost all of the fruit were infected (Fig. 1). In the fungicide-treated plot, the infection frequency increased during each interval between sprays; however, it was significantly reduced by subsequent spraying (Table 3). Propineb reduced the infection frequency from 9.7% (detected in the fruit bagged just before the application of propineb on May 25) to 2.5%. The infection frequency did not increase until June 12, before the application of azoxystrobin (Table 3). With the application of azoxystrobin on June 12, the infected fruit were completely

cured. However, the infection frequency increased over the 15 days after the application of azoxystrobin to reach 35.2% and was reduced to 10.5% by the application of iminoctadine-triacetate on June 26 (Table 3). The infection frequency increased again thereafter to reach 23.5% by July 11; however, this was also reduced to 7.5% by folpet (Table 3).

Although the proportion of disease incidence to infection frequency in the fungicide-untreated plot in 1999 was much higher than the previous two years (Fig. 1), the overall disease incidences in the fungicide-treated plot were too low to reasonably assess the after-infection activities of each of the fungicides. The rise and fall of disease incidence before and after each spray are shown in Table 3. The disease incidence of the fruit bagged just before the application of propineb on May 28 was 1.2%, which was completely reduced by the application of the fungicide (Table 3). No diseased fruit was found among those bagged before and after application of azoxystrobin. Disease incidence increased to 8.8%, along with a rapid increase of infection frequency, during the 14 days from June 12 to June 26, and this was completely reduced by iminoctadine-triacetate applied on June 26 (Table 3). The disease incidence increased again to 7.5% by July 11; however, it was reduced to 2.5% by the application of folpet on the same day (Table 3).

From the results of the trials over three years, it was confirmed that some fungicides, regardless of whether they were systemic or non-systemic, confer high after-infection

activity against white rot. Parker and Sutton (1993a) failed to detect post-infection activity in an experiment with five fungicides, benomyl, flusilazole, mancozeb, tebuconazole and triflumizole, by applying them on artificially inoculated apples. They determined the efficacy of the fungicides not by natural disease development, but by the growth of the fungus from an inoculated portion of apple tissue on culture medium. Despite the difference in experimental methods, benomyl and mancozeb also did not confer after-infection activities in our experiment. However, tebuconazole, for which Parker and Sutton could not detect after-infection activity, conferred very high after-infection activity when it was sprayed in early- or mid-August, curing most infected fruit regardless of the time of infection (Kim and Uhm, 2004).

In general, the after-infection activity or curative activity of fungicides is greater when they are applied as early as possible following the onset of infection (Solel, 1977). In this study, the period between infection and application of the fungicides that can confer after-infection activity was not determined. However, because some of the fruits exposed to natural inoculum for 14 to 16 days were cured, the period of time between infection and application of the fungicide might be relatively long in some fruits; a maximum of 14 to 16 days. This might be explained by the characteristic infection process of the white rot fungus. The main infection course of this fungus is via lenticels on the apples, which are composed of nonliving corked cells (Hayashi, 1984). Apple fruit can be infected early in the growing season through lenticels; however, the infections may remain latent during the immature stage of apple growth (Brown and Britton, 1990; Drake, 1971; Parker and Sutton, 1993b). The behavior of the fungus during the latent period is still not fully understood. Kim et al. (2001) reported that hyphae are commonly found between and within the subepidermal cells of immature apple tissue (cv. Fuji); however, unlike mature fruit, dissolution of cell walls was rarely found. Kim (2000) observed the proliferation of mycelia in and on the lenticels by laser confocal microscopy and scanning electron microscopy. Considering these two reports, the fungus probably grows in the nonliving cork tissue around the lenticel. The fungus growing in the lenticel might be killed or its growth might be suppressed by the fungicides that can easily soak into the nonliving corked cells. Therefore, the difference in after-infection activities observed by the four fungicides tested in this experiment might be caused by differences in the ability to kill or suppress activity against mycelia, permeability of the fungicide to corked cells, and other factors. However, why some fungicides can reduce the infection frequency, some reduce the disease incidence, and others reduce both, is still unclear.

It was repeatedly elucidated by the trials over two successive years from 1998 that the fungicide activities increased when the chemicals were sprayed alternately with other fungicides. Therefore, white rot could be controlled effectively by application of the chemicals showing after-infection activities because they would not permit the accumulation of infection. In addition to after-infection activities, white rot can also be effectively controlled, as previously mentioned, by the application of ergosterol biosynthesis inhibitors (EBIs), especially tebuconazole, in early- or mid-August because EBIs can greatly reduce the incidence of infection and disease of white rot, regardless of the infection time (Kim and Uhm, 2004). By applying these two facts, highly efficient fungicide programs of 15- and 25-day spray intervals have been developed. The process of development and improvement of the spray program will be published in subsequent papers.

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