

# Black Leaf Mold Development and Its Effect on Tomato Yield

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## ABSTRACT

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The severity of black leaf mold, caused by *Pseudocercospora fuligena*, was compared under epidemic conditions in field experiments on susceptible tomato lines that were either inoculated with the fungus or not inoculated. In one trial, 10 tomato entries that were infected by field inoculum had an average black leaf mold severity of 53 and 60% recorded over two assessments taken during fruit-set. In another experiment, different levels of disease severity were induced on a breeding line, CL 5915-153D<sub>4</sub>-3-3-0 (CL 5915) and a commercial variety, TN 2, by inoculating plants at various intervals. Both entries had up to 32% less yield than control plants that were kept disease-free with fungicides. CL 5915 had up to 11% and TN 2 up to 28% fewer fruit per plant than disease-free controls. Fruit weight was reduced to 20 and 7% of control plants for CL 5915 and TN 2, respectively. There was a significant negative correlation between the area under the disease progress curve and total yield, fruit number, and weight per fruit.

*Pseudocercospora fuligena* (Roldan) Dieghton causes black leaf mold of tomato (*Lycopersicon esculentum* Mill.) (12). The disease has been reported primarily from Asia (4,6-8,10,14,15), although there is one report from Florida (1). Lesions initially occur on lower leaves as chlorotic spots 1-5 mm wide. The infected areas turn light brown and then black as the fungus sporulates profusely. Heavily infected leaves wither and eventually drop from the plant.

There is limited information about black leaf mold and its causal agent. The disease and its pathogen were first described in the Philippines in 1938 (12), and the conditions most conducive for infection were studied (9). In the United States, the disease was first described in Florida in 1974 (1). In one report from Japan (15), the symptoms of the disease were described, the fungus was first grown in pure culture, varieties were assessed for resistance, and the survival of the fungus was determined on dried leaves kept in the laboratory. In Taiwan, the conditions for conidial germination and inoculation of tomato and *Solanum nigrum* L. were reported (5). There are no reports on how the disease develops in the field or how the disease affects yield. The objectives of our study were to assess and monitor black leaf mold under field conditions and to determine the relationship between disease parameters and yield components.

## MATERIALS AND METHODS

A regional yield trial for fresh market hybrid tomato consisting of eight test hybrids and two commercial hybrids (Taichung ASVEG #4 and Known You 301) was evaluated for naturally occurring black leaf mold at the Asian Vegetable Research and Development Center in Taiwan. Each tomato entry was transplanted on 28 August 1990 in four-row plots 5 m long. There was 0.75 m between rows and 0.5 m between plants in a row. Entries were arranged in a randomized complete block design with four replications. Black leaf mold was assessed twice during fruit-setting by estimating the percentage of leaf area infected on a per-plot basis on 21 December 1990 and 2 January 1991. Data were analyzed by ANOVA, and means were separated by LSD ( $P < 0.05$ ).

In a separate experiment that was repeated in two seasons (trials 1 and 2), seeds of a breeding line, CL 5915-153D<sub>4</sub>-3-3-0 (CL 5915), and a commercial variety, TN 2, were sown in flats in the greenhouse on 11 August 1989 and 18 September 1990. Forty-eight seedlings were transplanted in four rows, 40 cm apart, in two 1.5 × 5 m raised beds on 3 October 1989, and 24 seedlings were transplanted in two rows, 15 m apart, in two 1.5 × 5 m raised beds on 18 October 1990. The trials had split plots arranged in a randomized complete block design with four replications. Main plot treatments for trial 1 were as follows: treatment 1 = inoculated with *P. fuligena* on 3, 6, 14, 20, and 27 November and 4 December; treatment 2 = inoculated 27 and 29 November and 4, 12, and 15 December; treatment 3 = not inoculated; and treatment 4 = not inoculated and protected with benomyl (Benlate 50WP, 0.5 kg a.i./ha) and maneb (Dithane M-45, 1.6 kg a.i./ha) biweekly beginning on

17 October. In trial 2, main plot treatments were as follows: treatment 1 = inoculated on 23 October and 2, 6, 13, 15, 19, and 22 November; treatment 2 = inoculated on 18 November and 4 and 10 December, treatment 3 = inoculated on 13, 20, and 28 December; and treatment 4 = not inoculated and protected with fungicide (as described previously) beginning on 11 October. Subplots were tomato genotypes CL 5915 and TN 2 randomized within main plots.

Conidia of *P. fuligena* were harvested by adding 10 ml of distilled water to 10-day-old colonies grown on tomato leaf extract-oatmeal agar (5) and then rubbing the colonies with the end of a glass microscope slide to free conidia. The concentration of conidia was determined with a hemacytometer and adjusted to approximately 10<sup>3</sup> conidia per milliliter. Plants were sprayed until runoff with a hand-pump sprayer. Before inoculation, plants were irrigated overhead for approximately 10 min.

Disease was assessed 12 times at 7-day intervals from 23 November to 6 February in trial 1 and 11 times at 6- to 8-day intervals in trial 2. Disease severity was visually estimated by recording the percentage of leaf area infected after walking around the parameter of each plot. The values for the area under the disease progress curve (AUDPC) were calculated as described elsewhere (13).

Fruits were harvested from a 3 × 5 m area 11 times from 20 December to 28 February in trial 1 and six times from 24 December to 20 February in trial 3. Total fruit weight and the number of fruit per plot were recorded from each harvest and added over harvest dates. Individual fruit weight was calculated by dividing the total fruit weight by the number of fruit. A similar experiment was repeated on 3 April 1990. However, no fruits were harvested in this test because heavy rains led to loss in plant stand and a severe epidemic of bacterial spot. Data from this trial were not included in the results.

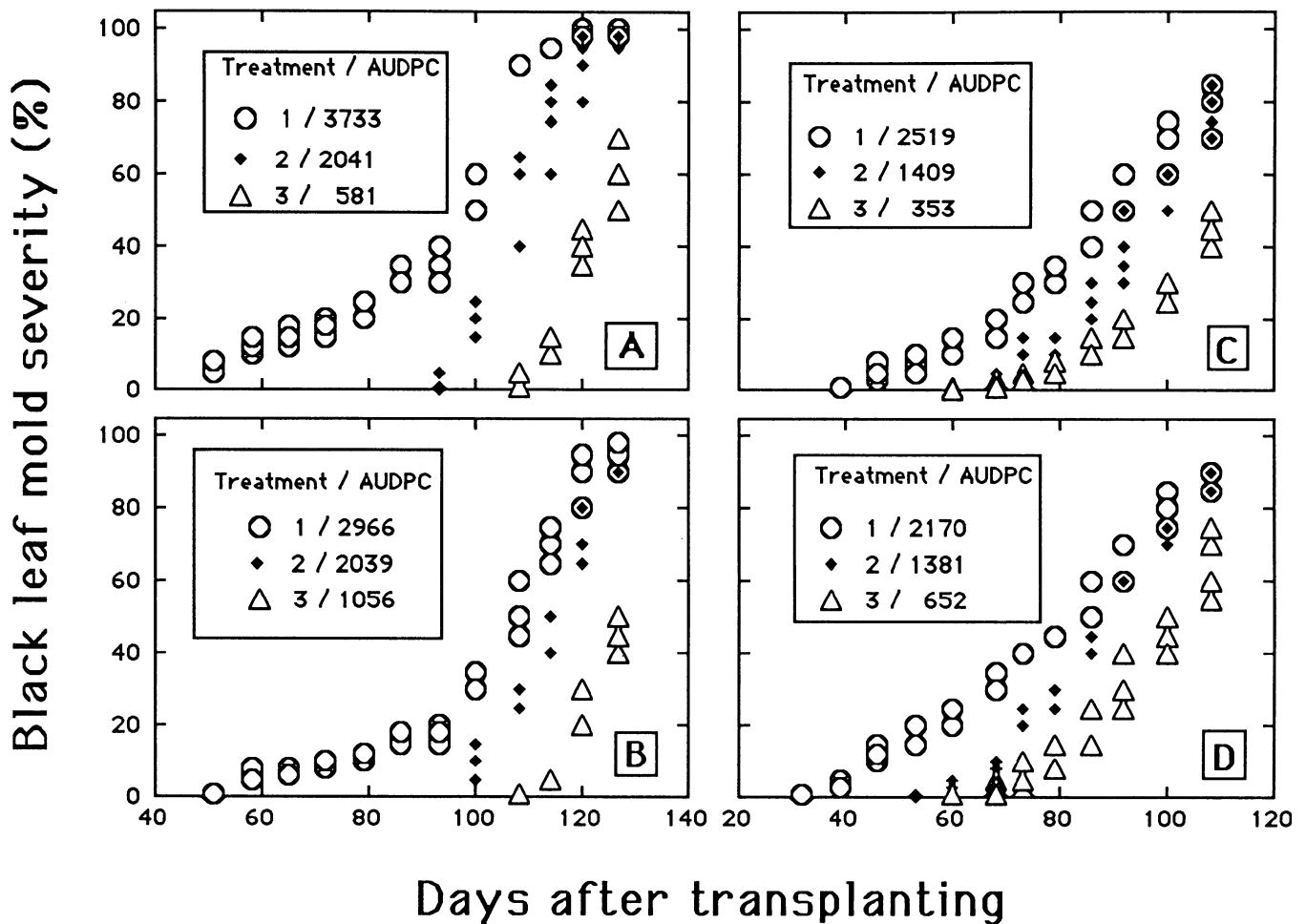
Data were analyzed by ANOVA, and means were separated by LSD ( $P < 0.05$ ). Regressions of yield components to AUDPC were calculated. Temperature, humidity, and rainfall data were collected daily during each experiment in an attempt to correlate these conditions with black mold severity.

## RESULTS

On 10 tomato entries, black leaf mold severity averaged 53 and 60% on the first

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**Fig. 1.** Development of black leaf mold and area under disease progress curve (AUDPC) on tomato lines (A and B) CL 5915-153D<sub>4</sub>-3-3-0 and (C and D) TN 2 in (A and C) trial 1 and (B and D) trial 2. In trial 1, treatments 1 and 2 were inoculated six and five times, respectively, with *Pseudocercospora fuligena*. Treatment 3 was not inoculated. In trial 2, treatments 1 and 2 were inoculated seven and four times, respectively. Treatment 3 was not inoculated.

and second assessment dates, respectively. Known You 301 had significantly more disease (75%) than the other entries in the first assessment, but there was no significant difference among entries in the second assessment. The lowest disease severity in the second assessment was 53% for both fresh market tropical tomato (FMTT) 274 and FMTT 305.

No disease occurred on foliage of CL 5915 or TN 2 foliage that were in non-inoculated and fungicide-protected plots. The later inoculation treatments were delayed, the more the disease progress curve was shifted to the right (Fig. 1). The disease increased slowly on plants inoculated early in the season, and the disease developed rapidly in plants that were inoculated later in the season. Within each treatment, black leaf mold severity increased more rapidly on CL 5915 than on TN 2, and there was a significant difference in the AUDPC values between the two tomato entries within each treatment (Table 1).

Total weight of harvested fruit was 4–32% (trial 1) and 12–34% (trial 2) less for plants with black leaf mold than for control plots (Table 2). Total fruit weight

**Table 1.** Area under disease progress curve of two tomato entries inoculated or not inoculated with *Pseudocercospora fuligena*

Treatment	Trial 1		Trial 2	
	CL 5915	TN 2	CL 5915	TN 2
Inoculated six to seven times <sup>a</sup>	3,733	2,519	2,966	2,170
Inoculated four to five times <sup>b</sup>	2,041	1,409	2,039	1,381
Not inoculated	581	353	1,056	652
LSD ( $P < 0.05$ ) <sup>c</sup>	134		139	
LSD ( $P < 0.05$ ) <sup>d</sup>	111		293	

<sup>a</sup>Six and seven times in trials 1 and 2, respectively.

<sup>b</sup>Five and four times in trials 1 and 2, respectively.

<sup>c</sup>Difference between means of tomato entries within the same treatment.

<sup>d</sup>Difference between means of tomato entries with different treatments.

averaged over treatments was not significantly different between CL 5915 (167 t/ha) and TN 2 (168 t/ha) in trial 1 or trial 2. Significant differences were detected in the number of fruit harvested among the inoculation treatments and between entries (Table 2). Fruit number was reduced from 0.1 to 19% for CL 5915 and from 8 to 28% for TN 2 compared with control plots. The weight per fruit was significantly less from plants in plots that were inoculated early in the season. Weight per fruit was 4–20% less than

control plots for CL 5915 and 0–13% for TN2.

As the AUDPC increased, the yield significantly decreased in both trials (Fig. 2). Predicted yields of the two lines decreased similarly. However, the number of fruits and their weights differed significantly between lines. Fruit production of CL 5915 did not decrease significantly as AUDPC increased, but the weight per fruit did decline significantly. Fruit production and the weight per fruit of TN 2 declined significantly as the

AUDPC increased.

During the experiments, the maximum relative humidity was more than 97%, temperatures were moderate and good for growth of tomato plants, and rainfall was <5 cm with less than 11 accumulative days of rainfall during two of the experiments (Table 3).

### DISCUSSION

Black leaf mold was first reported in Asia more than 50 yr ago, but there is little information on the importance of

this disease either under local conditions or over a wide geographical area. In our studies, the severity of black leaf mold exceeded 50% on all 10 tomato lines tested. This indicates there is a high degree of susceptibility in fresh-market hybrid tomatoes. The disease progressed slowly on younger inoculated plants but increased rapidly as plants aged. We also observed that the incubation period was fairly long and confirmed that symptoms do not develop until 10–14 days after inoculation (5,15). Although it appears

that disease buildup is slow after initial infection, the rate of disease development increases dramatically once the fungus begins to sporulate. The disease did not seem to be restricted by temperature within the normal range of tomato production or by the lack of rainfall (Table 3). It may be that free moisture actually limits black leaf mold. In an earlier study, conidia survived and germinated well below 100% relative humidity (5). More studies need to be conducted to determine how environmental factors influ-

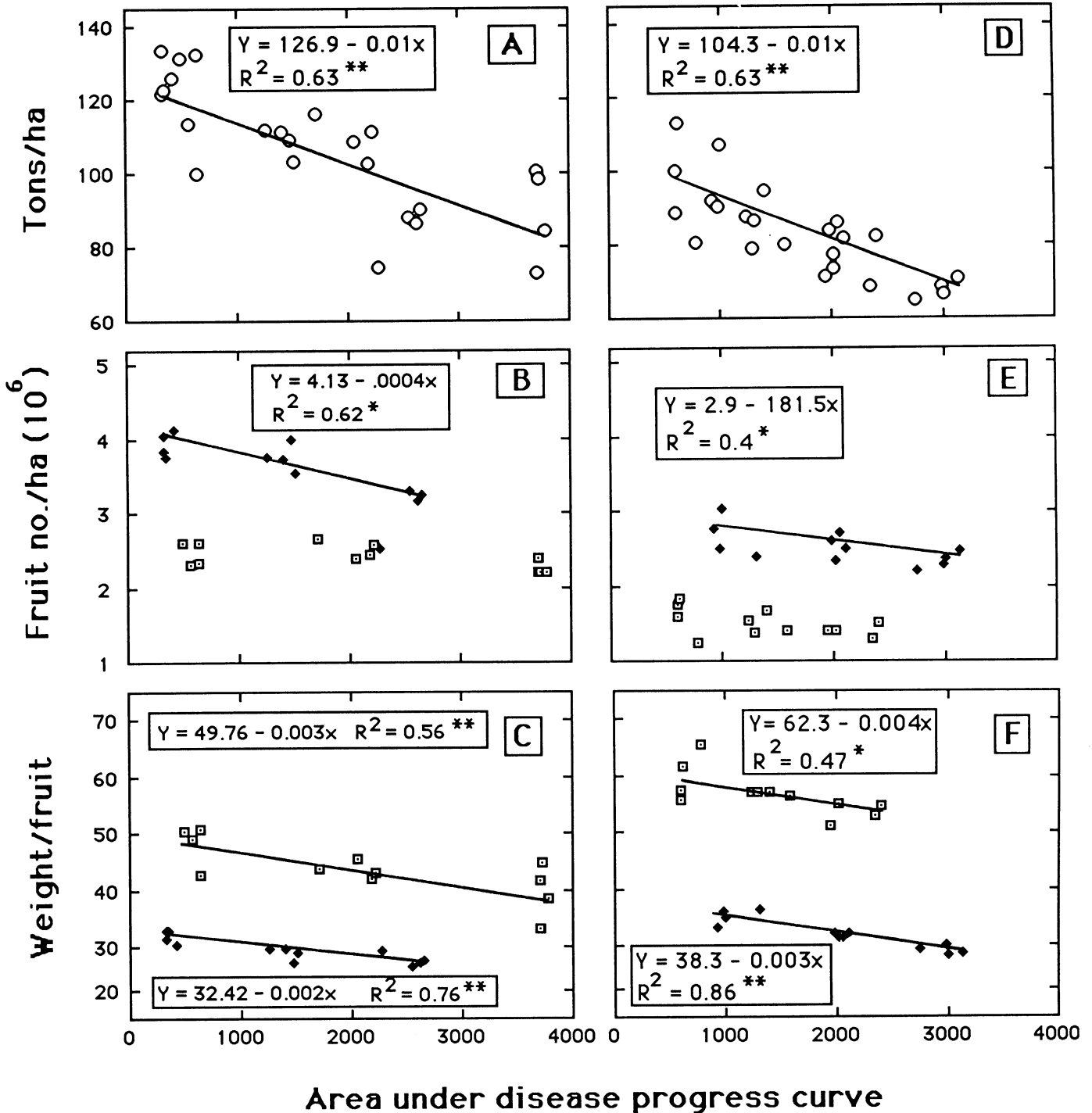


Fig. 2. Regression lines of tons per hectare (combined analysis of CL 5915-153D<sub>4</sub>-3-3-0 and TN 2), fruit number per hectare, and weight per fruit to area under disease progress curve for CL 5915-153D<sub>4</sub>-3-3-0 (◆) and TN 2 (□) infected with *Pseudocercospora fuligena* in (A-C) trial 1 and (D-F) trial 2. \* = Significant at  $P < 0.05$ , \*\* = significant at  $P < 0.01$ .

**Table 2.** Total fruit weight, fruit number, and weight per fruit of tomato lines CL 5915 and TN 2 in plots inoculated with *Pseudocercospora fuligena*, not inoculated, or fungicide-protected

Treatment	Total fruit weight		CL 5915		TN 2		CL 5915		TN 2	
	T/ha	Loss (%)	Fruit no.	Loss (%)	Fruit no.	Loss (%)	Wt./fruit	Loss (%)	Wt./fruit	Loss (%)
Trial 1										
Inoculated six times	130	32	3,369	11	4,587	28	40	20	28	7
Inoculated five times	164	15	3,772	0.1	5,620	12	44	12	29	3
Not inoculated	184	4	3,695	3	5,914	8	48	4	32	...
Fungicide-protected	192		3,800		6,403		50		30	
LSD ( $P < 0.05$ ) <sup>a</sup>	17									
LSD ( $P < 0.05$ ) <sup>b</sup>			684		684		3.9		3.9	
LSD ( $P < 0.05$ ) <sup>c</sup>			708		708		4.1		4.1	
Trial 2										
Inoculated seven times	71	34	1,399	19	2,329	27	54	16	29	13
Inoculated four times	83	23	1,496	12	2,538	20	57	11	32	5
Not inoculated	95	12	1,596	6	2,674	16	60	6	35	...
Fungicide-protected	109		1,737		3,197		64		34	
LSD ( $P < 0.05$ ) <sup>a</sup>	11									
LSD ( $P < 0.05$ ) <sup>b</sup>			220		220		3.8		3.8	
LSD ( $P < 0.05$ ) <sup>c</sup>			174		174		11.4		11.4	

<sup>a</sup> Difference between treatment means (t/ha).

<sup>b</sup> Difference between means (fruit number) of tomato entries within the same treatment.

<sup>c</sup> Difference between means (fruit number) of tomato entries with different treatments.

**Table 3.** Range and mean relative humidity, temperature, and rainfall in three tomato field trials at the Asian Vegetable Research and Development Center in Taiwan

Dates of experiment	Relative humidity				Temperature				Rainfall	
	Maximum (%)		Minimum (%)		Maximum (C)		Minimum (C)		Cm	Days <sup>a</sup>
	Range	Mean	Range	Mean	Range	Mean	Range	Mean		
3 Oct. 1989–6 Feb. 1990	92–100	99	42–96	66	12–32	25	8–24	15	2	11
28 Aug. 1990–2 Jan. 1991	90–100	97	54–94	69	18–33	28	10–26	19	13	31
18 Oct. 1990–28 Feb. 1991	92–100	97	54–96	66	16–31	25	10–22	15	5	7

<sup>a</sup> Number of days that precipitation was recorded.

ence disease development.

Yield loss attributable to black leaf mold may not be as great as other tomato foliar diseases such as bacterial spot, early and late blight, and Septoria leaf spot. Losses of marketable fruit attributable to bacterial spot were as high as 53% with no apparent reduction in total fruit number (11). Early blight in non-inoculated plots was reported to reduce yields by 46% (2). In Nigeria, yield reductions attributable to Septoria leaf spot averaged 25, 41, and 52% at three locations (3). In general, reports dealing with yield losses of tomatoes caused by foliar pathogens have not documented whether reductions in fruit weight, total fruit number, or a combination of both is most important. Our studies with black leaf mold indicated that both fruit weight and number can be reduced, however, responses may vary among tomato lines. Yield losses attributable to black leaf mold may be lower than other foliar diseases because actual defoliation is delayed. Additionally, the disease seems to develop slowly, becoming severe only later in the season. Marketability is not affected because *P. fuligena* does not seem to cause direct damage to fruits. Defoliation can be especially critical because it increases sunscald of fruit, which also

was reported to be an important effect of bacterial spot (11). During our studies, we abandoned a trial because of a period of heavy rains, which was followed by a severe outbreak of bacterial spot. Black leaf mold never really developed, as there was little sporulation even though there was some initial infection. As bacterial spot increased, defoliation was so severe that there was little tissue left to be infected by *P. fuligena*.

Black leaf mold occurs in many Southeast Asian countries, and it has been reported to be important in Japan (15). It has not yet been shown to be economically damaging on tomato over a wide geographic region, possibly because its importance is not yet fully known. However, it is likely that more detailed reports will be forthcoming from other countries about the importance of this disease.

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