Evaluation of Resistance to Bacterial Wilt in Eggplant

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ABSTRACT

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Eggplant (*Solanum melongena*) accessions from the U.S. Plant Introduction Collection were evaluated in a greenhouse for resistance to bacterial wilt caused by *Pseudomonas solanacearum*. A few of the most resistant accessions were crossed with each other. From six of these crosses, F_1 and F_2 populations were developed. A mixture of three strains of *P. solanacearum* race 1 was used to inoculate 21-day-old seedlings by root dipping. After 3 wk, the surviving plants were stem-inoculated with a mixture of these same three strains. A genetic advance in the level of resistance to bacterial wilt was observed in the F_1 and F_2 progenies of three crosses: PI 176761 × PI 169663, PI 176761 × PI 320505, and PI 220120 × PI 173106.

Bacterial wilt caused by *Pseudomonas solanacearum* E. F. Smith is an important disease of eggplant (*Solanum melongena* L.) worldwide (5). The bacteria can survive in soils in areas of mild to warm climates (5,8,12) and can be the limiting factor for growing this crop. In North Carolina, field losses of 50% have been observed by midsummer (17), and a loss between 75 and 81% has been recorded in India (13).

Resistance to bacterial wilt has been studied in several crops, especially tomato (1,2,9,10,14), but there is little published work on resistance in eggplant. Eggplant cultivars have been evaluated for resistance to bacterial wilt in Puerto Rico (11), and breeding programs for bacterial wilt resistance have been conducted in Puerto Rico, the Philippines, Sri Lanka, Malaysia, India, and South Africa (15). In North Carolina, resistant eggplant cultivars such as Kopek from Java and Matale from Sri Lanka have been tested, but neither has produced marketable fruit (17).

In 1985, all 524 eggplant accessions in the U.S. Department of Agriculture's Plant Introduction Collection were evaluated for resistance to *P. solanacearum* (4). Although none of the accessions was immune, some were quite resistant to *P. solanacearum*. Several of the most resistant lines were crossed with each other in an attempt to combine genes that might improve resistance levels. From six of these crosses, F_1 and F_2 populations were developed and then evaluated for bacterial wilt resistance.

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MATERIALS AND METHODS

In the winter of 1986–1987, six crosses with their F_1 and F_2 populations, as well as two susceptible commercial cultivars, Round Oval Hybrid and Viserba, were tested for resistance to bacterial wilt. Plants were sown in 1-in. Styrofoam Speedling flats (Speedling, Inc., Sun City, FL) containing Jiffy Mix Plus (Jiffy Products of America, West Chicago, IL) and were grown in a greenhouse. Night temperatures fluctuated between 24 and 27 C and daytime temperatures, between 33 and 37 C.

P. solanacearum strains GA_2 and AL_2 (supplied by S. M. McCarter) and G-4 (from R. W. Goth) were maintained in capped test tubes of 5 ml of sterile distilled water at room temperature to reduce development of avirulent mutants (3,7). Inoculum for this study was produced by growing the strains in nutrient broth cultures (3,6,7). Three-day-old broth cultures were used directly as inoculum after adjusting the bacterial concentration. Klett units with a Klett-Summerson photoelectric calorimeter (green filter) coupled with a 10-fold dilution series were used to adjust the concentration of the broth inoculum to 10^8 cfu/ml.

When 21 days old, seedlings were carefully lifted from the flats. The roots were washed in tap water, then dip-inoculated with the mixture of the three strains. Two checks, Round Oval Hybrid and Viserba, were dipped into the suspension of the three strains and, as a negative control, into nutrient broth. The

inoculated seedlings were immediately transplanted in a pasteurized soil mix in 10-cm-diameter pots and kept in a greenhouse. There were three replications each of 24 plants for parents and the F_1 population and of about 128 plants for the F_2 population. There were 12 plants for each treatment of the checks.

After inoculation, soil moisture was maintained at a high level by watering the plants heavily four times a day. Temperatures were maintained between 24 and 27 C at night and 33 and 37 C during the day. Low-pressure sodium lights (Norelco SOX 180W) were used to increase day length to 14 hr.

The inoculated plants were observed daily, and records were kept of the date of appearance of wilt symptoms and of plant death. From these data, we calculated percent disease at the end of the test and the number of days when 50% of the plants were wilted. Twenty days after the root-dip inoculation (the first inoculation), survivors of more than five plants in each line were inoculated (the second inoculation) with the same bacterial strains by the stem-puncture methods of Winstead and Kelman (16). For this second inoculation, disease readings (no symptoms = 0, one leaf wilted = 1, two or three leaves wilted = 2, all leaves wilted except top two or three = 3, all leaves wilted = 4, dead = 5) were taken at 7-day intervals and converted to disease indices (16). Diseased plants were often examined for vascular discoloration, a characteristic of invasion by *P. solanacearum*, and reisolations made from them. Data were analyzed by analysis of variance and Duncan's new multiple range tests.

RESULTS

Under these test conditions, none of the eggplant PI lines showed immunity or a high level of resistance to bacterial wilt. However, some lines and their progeny showed various levels of resistance (Table 1). PI 230334 and PI 320502 in cross 5 and PI 349612 in cross 6 had a disease incidence of about 40%, showed delayed disease development in the root-dip inoculation, and had a disease index of less than 30 in the second inoculation. By contrast, more than 80% of the plants of Round Oval Hybrid and Viserba and of the PI lines 173106, 174367, and 220120 and populations derived from them in crosses 1 and 4 became diseased. These plants wilted and died rapidly.

In cross 6, PI 349612 had less disease than the other parent, PI 173106, and resistance from PI 349612 was transmitted to the F_1 and F_2 generations. In cross 3, PI 320505 imparted some resistance to progeny when crossed with the more susceptible PI 176761.

There was a marked genetic advance in the level of resistance to bacterial wilt in the F_1 and F_2 generations from the cross of some parents. In cross 2, disease incidence was lower and disease development was delayed in the first inoculation and the disease index was lower in the second inoculation in the F_1 and F_2 generations than in either parent, PI 176761 and PI 169663. In addition, after the second inoculation, both F_2 populations in crosses 3 and 4 had much higher resistance than the two parents.

Table 1. Bacterial incidence and severity in eggplant lines infected with *Pseudomonas solanacearum* under greenhouse conditions

Cross no.	Population		Root-dip (first) inoculation[x] Percent diseased	Root-dip (first) inoculation[x] Days to 50% of plants wilted	Stem-puncture (second) inoculation[y] Disease index
1	P_1	PI 174367	83.7 a[z]	7.0 a	41.0 c
	P_2	PI 173106	88.7 a	7.3 a	66.5 a
	F_1		77.9 a	7.5 a	65.7 ab
	F_2		82.5 a	6.9 a	52.7 bc
2	P_1	PI 176761	74.9 a	9.8 ab	58.0 b
	P_2	PI 169663	74.9 a	7.2 b	84.0 a
	F_1		48.7 b	15.0 a	43.4 c
	F_2		45.5 b	15.3 a	39.8 c
3	P_1	PI 176761	74.9 a	9.8 a	58.0 a
	P_2	PI 320505	52.3 b	13.7 a	46.7 b
	F_1		61.1 ab	11.1 a	45.8 b
	F_2		62.5 ab	13.2 a	33.8 c
4	P_1	PI 220120	90.3 a	5.4 a	77.0 a
	P_2	PI 173106	88.7 ab	7.3 a	66.5 ab
	F_1		87.6 ab	7.2 a	57.6 b
	F_2		70.2 b	9.2 a	32.5 c
5	P_1	PI 230334	39.1 a	18.3 a	26.5 a
	P_2	PI 320502	40.4 a	16.0 a	23.5 a
	F_1		48.3 a	15.4 a	22.5 a
	F_2		39.5 a	18.3 a	21.7 a
6	P_1	PI 349612	35.5 b	20.0 a	11.7 b
	P_2	PI 173106	88.7 a	7.3 b	66.5 a
	F_1		41.5 b	20.0 a	18.0 b
	F_2		47.5 b	20.0 a	21.0 b
Control	Round Oval Hybrid		89.0 a	6.8 a	85.1 a
	Viserba		65.3 a	8.3 a	97.1 a

[x] This portion of the test was terminated 20 days after inoculation.
[y] Survivors of the root-dip inoculation were inoculated by stem puncture, and disease indices were calculated after 20 days.
[z] Within each cross, means followed by the same letter are not significantly different at the 5% level by Duncan's new multiple range test.

DISCUSSION

No immunity was found to *P. solanacearum* in the cultivated eggplant species *S. melongena*. Both types of inoculations performed are very severe screening tests and are responsible for identifying a few eggplant PI lines that can be used as sources of resistance.

There was little difference in the resistance found in some PI lines between this test and the screening test reported previously (4). In both tests, PI 349612 showed relatively high resistance. PI 176761, however, was susceptible in this test but not in the previous one. This variation could be caused by the use of two additional and different bacterial strains or by the higher inoculum concentration used in this test. Moreover, in an additional very severe screening test of 23 eggplant PI lines in a greenhouse, PI 176761 was highly susceptible to bacterial wilt, whereas PI 349612 was moderately resistant (*unpublished*).

Little information has been reported on the mode of inheritance of resistance to bacterial wilt in eggplant. We demonstrated that a low level of resistance can be transmitted to progeny from some eggplant PI lines. In cross 6, where the F_1 does not differ significantly from the more resistant parent, dominance is shown. In cross 3, where the F_1 is midway between the parents, no apparent dominance is shown. In cross 2, resistance was improved by combining genes from two selected lines, and this may be an example of minor gene additive effects where some genes are contributed by both parents. The complex nature of wilt resistance in eggplant appears to be similar to reports showing polygenes for resistance in tomato or a partially dominant or recessive inheritance of tomato bacterial wilt resistance (1,2,14).

Use of this genetic material should be beneficial in gradually improving the level of resistance of eggplant to bacterial wilt. The gradual shift toward resistance

may eventually be used to delay the onset of wilt symptoms and could result in germ plasm that can be used to produce an economical and marketable yield.

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