Differential Susceptibility of *Malus* spp. Cultivars Robusta 5, Novole, and Ottawa 523 to *Erwinia amylovora*

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ABSTRACT

Norelli, J. L., Aldwinckle, H. S., and Beer, S. V. 1986. Differential susceptibility of *Malus* spp. cultivars Robusta 5, Novole, and Ottawa 523 to *Erwinia amylovora*. Plant Disease 70:1017-1019.

Twenty-five apple cultivars were inoculated with two strains of *E. amylovora* to evaluate their fire blight susceptibility. Three cultivars were highly resistant to strain Ea 273 but were susceptible to strain Ea 266. These included Robusta 5 and Novole, which were previously identified as fire blight-resistant, and Ottawa 523. The data indicate these cultivars are differentially susceptible to strain Ea 266.

Differential host × pathogen interactions were recently described among cultivars of apple (*Malus* spp.) and strains of *Erwinia amylovora* (Burr.) Winslow et al (6), the incitant of fire blight. Inoculation of shoots of most apple cultivars with *E. amylovora* strains Ea 273 and Ea 266 resulted in similar amounts of disease. However, the cultivar Quinte was differentially susceptible to strain Ea 266.

Gardner et al (2) reported certain cultivars of *Malus* spp. are highly resistant to fire blight. The resistant cultivars, identified by inoculation with *E. amylovora* strain Ea 273, include *M.* × *atrosanguinea* Has. 24, Robusta 5 (*M.* × *robusta* No. 5), and Novole (*M.* × *sublobata* PI 286613).

The purpose of this study was to compare the susceptibility of several apple cultivars to *E. amylovora* strains Ea 273 and Ea 266. The cultivars of apple studied included commercial cultivars, breeding selections, and cultivars of *Malus* spp. Some of the cultivars previously reported by Gardner et al (2) as highly resistant to fire blight were susceptible to *E. amylovora* strain Ea 266.

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MATERIALS AND METHODS

Bacterial strains and inoculum. *E. amylovora* strain Ea 273 originated in Wayne County, New York. Strain Ea 266 originated in Simcoe, Ontario, Canada, and was originally isolated as strain E4001A. Both strains of *E. amylovora* were isolated from the same host, Rhode Island Greening apple. Strains Ea 273 and Ea 266 are maintained in the Cornell University Collection of Phytopathogenic Bacteria as strains 0273 and 0071, respectively.

Broth cultures used for inoculation were started from single colonies of freshly revived lyophilized cultures grown on nutrient broth/yeast extract/glucose agar. Inoculum consisted of 18- to 24-hr-old cultures grown in Kado 523 broth (4) at 28 C and contained 10^9-10^{10} colony-forming units per milliliter.

Plant material. The susceptibility of selected apple cultivars was compared in greenhouse tests. Trees were grown from bench grafts on seedling rootstocks in the greenhouse and trained to single shoots by methods described previously (1).

Inoculation and disease measurement. Trees with vigorously growing vegetative shoots at least 20 cm long were selected and placed randomly in two groups for inoculation with either strain. A mean of 12 (eight to 22) trees were inoculated per cultivar, depending on availability. Shoot tips were inoculated on 16 June 1984 by inserting a 0.46-mm-diameter (26-gauge) hypodermic needle through the stem just above the youngest unfolded leaf. Enough inoculum was introduced to fill the wound and leave visible drops at both ends of the wound. Greenhouse temperatures ranged from about 15 to 33 C during disease development. The lengths of visible fire blight lesions and of current season's shoot growth were recorded after all lesions had ceased to extend, as determined by the formation of a determinate margin between diseased and healthy tissue. The mean proportion of the current season's shoot length that was necrotic was calculated and considered a measure of susceptibility. Lesions that extended into the previous season's growth were rated as 1.00. Differences in the virulence of strains, susceptibility of cultivars, and interactions among cultivars and strains were analyzed by a two-way analysis of variance after angular transformation of data. Multiple comparisons of cultivar susceptibility were performed separately for each strain by Waller and Duncan's

Bayesian *K*-ratio procedure (8) after angular transformation of the data.

RESULTS AND DISCUSSION

Apple cultivars Robusta 5, Ottawa 523, and Novole were highly resistant to *E. amylovora* strain Ea 273 but susceptible to strain Ea 266 (Table 1). Results indicated this interaction was due to the differential susceptibility of these cultivars to strain Ea 266. Results of a two-way analysis of variance indicated a significant strain × cultivar interaction at $P = 0.01$ (Table 1). In addition, the rankings of cultivar susceptibility differed when the cultivars were inoculated with strains Ea 273 and Ea 266. Both Robusta 5 and Ottawa 523 were ranked significantly less susceptible to infection than Haralson when inoculated with strain Ea 273, but neither ranked significantly different from Haralson when inoculated with Ea 266. Similarly, Robusta 5 ranked significantly less susceptible than Delicious when inoculated with strain Ea 273 but significantly more susceptible than Delicious when inoculated with strain Ea 266. A significant strain × cultivar interaction in a two-way analysis of variance and a change in susceptibility ranking with different strains are both indications of differential host × pathogen interactions (7).

Strain Ea 266 was generally more aggressive (7) than Ea 273, with overall mean disease severity values of 0.93 and 0.63, respectively. However, increased aggressiveness does not explain the reactions of some cultivars to inoculation with strain Ea 266. The disease severity values that resulted when cultivars were inoculated with strain Ea 273 were strongly correlated ($r = 0.835$, df = 23) with the values from cultivars inoculated with Ea 266 (Fig. 1). However, the cultivars Ottawa 523, Robusta 5, and State Fair were outliers and did not fit this relationship (Fig. 1). When these three cultivars were removed from the correlation analysis, the correlation coeffecient (r) increased to 0.943, df = 20.

The differential susceptibility of Novole to strain Ea 266 is less evident from the data than that of Robusta 5 and Ottawa 523. The susceptibility ranking of Novole was not significantly different when the cultivars were inoculated with strain Ea 273 or with Ea 266 (Table 1). However, several inoculation trials have indicated Novole is highly resistant to strain Ea 273 but susceptible to strain Ea 266. Naturally occurring fire blight has been observed on orchard trees of Robusta 5 in Geneva, NY, but not on Novole trees growing within the same block (J. L. Norelli, *personal observation*). This probably indicates a higher level of general resistance in Novole than in Robusta 5, as suggested by artificial inoculation data (Table 1).

The *M.* × *atrosanguinea* clone used in this experiment did not appear to be differentially susceptible to strain Ea 266. This cultivar is an undesignated clone obtained from R. D. Way, Department of Horticultural Sciences, Geneva, NY. Previous greenhouse tests of *M.* × *atrosanguinea* Has. 24, rated highly resistant by Gardner et al (2), indicated it was differentially susceptible to strain Ea 266. Ottawa 523, which appeared differentially susceptible to strain Ea 266 in this study, has *M.* × *atrosanguinea* in its parentage (Red Melba × [Wolf River × *M.* × *atrosanguinea* 804]). Because differential host × pathogen interactions occur among cultivars of apples and strains of *E. amylovora* (6), it is not surprising that different genotypes of *M.* × *atrosanguinea* vary in their susceptibility

Table 1. Disease severity values[w] of fire blight on apple cultivars inoculated with *Erwinia amylovora* strains Ea 273 and Ea 266 and summary of analysis of variance

Cultivar	Ea 273	Ea 266
State Fair	1.41 a[x]	1.23 abcde
Rome Beauty	1.34 a	1.52 a
Discovery	1.21 ab	1.53 a
Summer Treat (NJ49)	1.17 ab	1.30 abc
Laura Red	0.98 bc	1.23 abcde
McIntosh	0.93 bc	1.17 bcde
NY 70707-92[y]	0.87 bcd	1.29 abcd
Golden Delicious	0.87 cd	1.36 abc
Haralson	0.86 cd	1.04 def
Jonathan	0.86 cd	1.39 a
NY 72702-60	0.82 cde	0.88 efgh
C48 interstem	0.75 cdef	0.85 fgh
Jonared	0.70 cdef	1.05 cdef
C42 interstem	0.66 cdef	0.90 efgh
C57 interstem	0.66 cdef	0.75 fgh
Aomori Red Delicious	0.62 cdef	0.65 ghi
Redfree	0.62 def	0.73 fgh
Delicious	0.57 ef	0.62 hi
Red Bouquet Delicious	0.41 fg	0.64 ghi
Liberty	0.26 gh	0.35 i
NY 58533-4	0.13 gh	0.31 ij
M. × *atrosanguinea*	0.02 h	0.03 j
Novole	0.00 h	0.33 ij
Ottawa 523	0.00 h	0.70 fgh
Robusta 5	0.00 h	0.93 efg
Strain mean	0.69	0.93

Source of variation	df	MS	F
Strains	1	4.054	···
Cultivars	24	1.801	···
Strains × cultivars	24	0.154	1.86[z]
Error	250	0.083	···

[w] Disease severity value was calculated from the arc sine $\sqrt{x}$, where x = the proportion of the current season's shoot length that was necrotic.

[x] Cultivars within a column followed by the same letter did not differ significantly at $P = 0.05$ according to Waller and Duncan's Bayesian *K*-ratio LSD rule (8).

[y] NY numbers are selections from the apple sicon breeding program of R. C. Lamb and H. S. Aldwinckle, Cornell University, Geneva, NY.

[z] Critical *F* values: $F0.05$ (df = 24,250) = 1.56; $F0.01$ (df = 24,250) = 1.87.

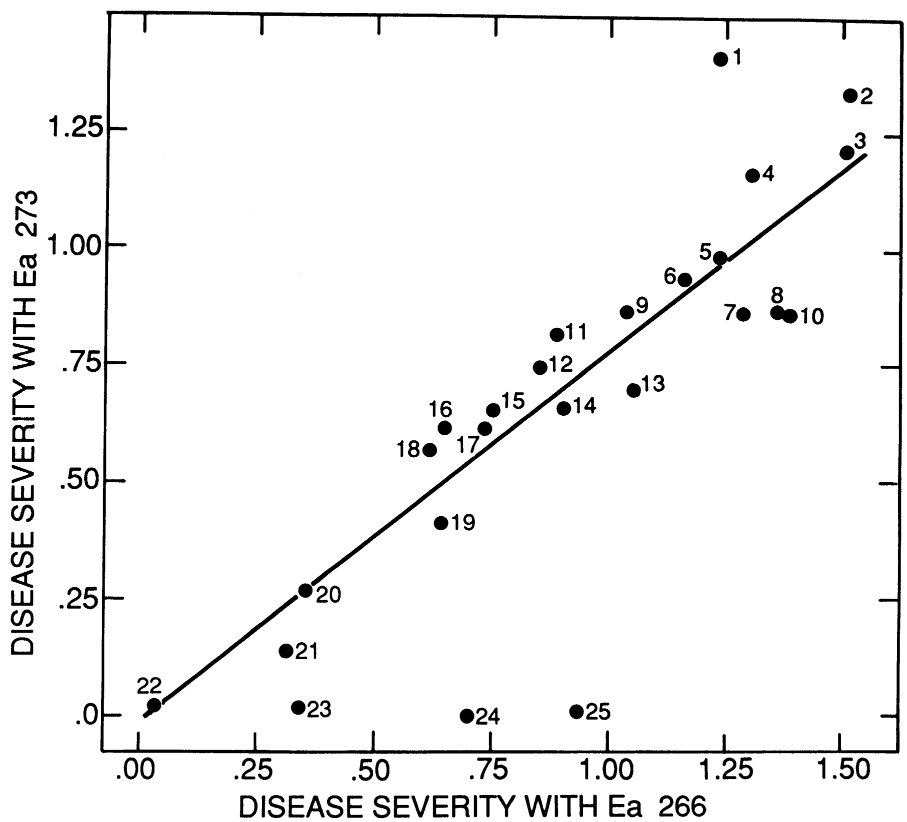

Fig. 1. Relationship between severity of fire blight on apple cultivars inoculated with *Erwinia amylovora* strains Ea 273 and Ea 266. Numbered points represent cultivars: 1 = State Fair, 2 = Rome Beauty, 3 = Discovery, 4 = Summer Treat (NJ49), 5 = Laura Red, 6 = McIntosh, 7 = NY 70707-92, 8 = Golden Delicious, 9 = Haralson, 10 = Jonathan, 11 = NY 72702-60, 12 = C48 interstem, 13 = Jonared, 14 = C42 interstem, 15 = C57 interstem, 16 = Aomori Red Delicious, 17 = Redfree, 18 = Delicious, 19 = Red Bouquet Delicious, 20 = Liberty, 21 = NY 58553-4, 22 = *M.* × *atrosanguinea*, 23 = Novole, 24 = Ottawa 523, and 25 = Robusta 5. The disease severity value was calculated from the arc sine $\sqrt{x}$, where x = the proportion of the current season's shoot length that was necrotic. The calculated regression line, $y = 0.8\,x - 0.0274$ ($r^2 = 0.89$), did not include points 1, 24, and 25 (State Fair, Ottawa 523, and Robusta 5, respectively).

to strains of *E. amylovora*.

Gardner et al (3) observed two types of fire blight resistance in apples. In commercial apple cultivars and some species clones, resistance is polygenically (quantitatively) controlled. The resistance of Robusta 5 and Novole is under oligogenic control (few dominant genes) (3). Gardner et al preferred oligogenic fire blight resistance for use in their *Malus* breeding program because of its ease of manipulation. However, based on the differential susceptibility of Novole and Robusta 5 to Ea 266, the oligogenic resistance of these cultivars may be less stable than polygenic resistance. The differential susceptibility of these cultivars is not a unique interaction with strain Ea 266. In a survey of more than 100 *E. amylovora* strains, Novole was differentially susceptible to about 10% of the strains tested (J. L. Norelli, *unpublished*). The differentially virulent strains identified were originally isolated in Michigan, Missouri, Nebraska, New York, and Ontario.

The disease-resistant apple cultivars, Liberty (5) and NY 58553-4, were partially resistant (not immune) when inoculated with either strain Ea 273 or Ea 266 in this study. The fire blight resistance of these cultivars is presumably the result of genes derived from *M. floribunda* 821, their source of resistance to *Venturia inaequalis* (5). Under commercial conditions, complete immunity to fire blight is not necessary for either apple scion or rootstock cultivars, because major economic losses are a function of severity of infection (extent of disease development) and not incidence of infection (proportion of blossoms or shoot tips infected). Liberty's fire blight resistance will be commercially useful and may be more durable than the oligogenic resistance of cultivars such as Robusta 5 and Ottawa 523.

Our results indicate that the evaluation and selection of fire blight resistance may be affected by the virulence of the strains of *E. amylovora* used in screening. In resistance breeding, steps must be taken to ensure that progenies are challenged by strains representative of the complete range of differential virulence in *E. amylovora*.

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