

Aggressiveness of Strains of *Pseudomonas solanacearum* from the French West Indies (Martinique and Guadeloupe) on Tomato

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ABSTRACT

Prior, P., Steva, H., and Cadet, P. 1990. Aggressiveness of strains of *Pseudomonas solanacearum* from the French West Indies (Martinique and Guadeloupe) on tomato. *Plant Dis.* 74:962-965.

Strains of *Pseudomonas solanacearum* indigenous to the French West Indies were divided into four profiles of aggressiveness on tomato cultivars Floradel, Capitan, and Caraïbo, which are susceptible, moderately resistant, and resistant, respectively. Depending on the strain used, the resistance expressed in Capitan was dependent on temperature, whereas that in Caraïbo and Floradel was not. A link appears to exist between aggressiveness in Capitan and biovar type.

Bacterial wilt, caused by *Pseudomonas solanacearum* (Smith) Smith, is very damaging to solanaceous crops in tropical and subtropical regions of the world (2,9). In the French West Indies (FWI), *P. solanacearum* limits the development and production of tomato (*Lycopersicon esculentum* Mill.) and eggplant (*Solanum melongena* L. [6]). Most strains of *P. solanacearum* belong to race 1 (3) and biovars I and III (7), but in Guadeloupe, race 3 was found in a cooler potato-growing area (15). Based on pathogenicity, there are at least five groups of strains with a homogeneous geographical distribution (15). Martinique and Guadeloupe breeding programs for resistance to bacterial wilt may use identical strains in terms of virulence (qualitative term) but entirely different ones in terms of aggressiveness or the quantitative variation of virulence (17). Such differences could account for reports of important losses in eggplant crops from Martinique but not from Guadeloupe (13), even though strains in the two islands belong to the same race and biovar.

The purpose of this study was to determine the variability of aggressiveness

among strains from both islands and areas within each island. The variation was evaluated within tomato cultivars rated as susceptible, moderately resistant, and resistant.

MATERIALS AND METHODS

Cultures. All 22 strains of *P. solanacearum* collected in the FWI were virulent on tomato (15) and have been deposited in the Collection Nationale des Bactéries Phytopathogènes (CNBP), INRA, Station de Pathologie Végétale et de Phytobactériologie, 49000, Angers, France (Table 1). Several strains (1000, K60, K74, K105, K136, S225, S236, and S247) obtained from the collection at the Department of Plant Pathology, University of Wisconsin, Madison, were included as controls. Cultures stored in sterile tap water in screw-cap tubes at room temperature were streaked on Kelman's tetrazolium chloride medium (TZC; 10). Fluidal, wild-type colonies were selected and restreaked on the same medium without TZC. After 48 hr at 28 C, the plates were flooded with sterile, distilled water, the colonies were suspended, and the suspensions were diluted to 10^7 cfu/ml measured spectrophotometrically ($A_{(650)} = 0.1$).

Plants. Seeds of three tomato cultivars (Floradel, Capitan, and Caraïbo) that differed in resistance to bacterial wilt (susceptible, moderately resistant, and

resistant, respectively [4]) were planted in disinfected vegetal soil. Each seedling was transplanted into a 10-cm pot and grown in a greenhouse at 27 ± 4 C. Plants were watered daily with tap water.

Inoculation. One milliliter of inoculum was poured around the base of 25-day-old plants (20–25 cm in height) and a knife was inserted 4–5 cm into the soil to cut the roots along one side (19). Inoculated plants were incubated in growth chambers at 32, 28, or 25 ± 2 C which represent very favorable, favorable, or weakly favorable temperatures for disease development. Plants received supplemental lighting with a 12-hr photoperiod and were watered daily with tap water. There were 10 plants for each combination of cultivar, strain, and temperature. Symptom development was recorded up to 21 days after inoculation, and the test was repeated with the two lower temperatures, 25 and 28 C.

Data analysis. A factorial correspondence analysis (FCA [1,8]) was carried out with the use of a table made up of 15 columns and 30 rows (Table 2). The columns correspond to the dependent number of symptomless plants of each cultivar after inoculation at each temperature. The rows correspond to the strains of the bacterium. In the FCA, each strain was represented by a point in space with as many dimensions as the number of combinations of cultivar and temperature; each combination of temperature and cultivar was a point in a space with as many dimensions as strains. As rows and columns are symmetrically analyzed, the points can be represented on the same plane, or for more convenience, on two different superimposing planes. For each space defined by strains or cultivar \times temperature combinations, orthogonal axes were calculated as linear combinations of the initial temperature and cultivar. The new parameters created by the analysis displayed the similarity

Accepted for publication 5 December 1989.

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Table 1. Strains of *Pseudomonas solanacearum* used in aggressiveness tests on tomato

Strains ^a	Host plant	Location	Year isolated	Race ^b	Biovar ^b
MT1	Tomato	Basse Pointe	1986	1	III
MT2	Tomato	Lamentin	1983	1	III
MT4	Tomato	Lamentin	1985	1	III
MT5	Tomato	Case Pilote	1987	1	I
GT1	Tomato	Saint-François	1985	1	I
GT4	Tomato	Petit-Bourg	1984	1	III
GT5	Tomato	Saint-Claude	1984	3	III
MA1	Eggplant	Morne L'Etoile	1985	1	III
MA2	Eggplant	Case Pilote	1984	1	III
MA3	Eggplant	Morne Vert	1985	1	III
GA1	Eggplant	Petit-Bourg	1984	1	III
GA2	Eggplant	Sainte-Rose	1986	1	III
GA3	Eggplant	Matouba	1984	1	III
GA4	Eggplant	Vieux-Habitants	1983	1	III
GA5	Eggplant	Saint-François	1983	1	III
MP1	Pepper	Fort de France	1985	1	III
GP1	Pepper	Petit-Bourg	1984	1	III
ME1	Potato	Lamentin	1986	1	I
ME2	Potato	Carbet	1987	1	III
ME3	Potato	Morne Vert	1987	1	I
GE1	Potato	Petit-Bourg	1987	1	I
MB1	<i>E. ventricosum</i>	Lamentin	1987	1	III
Reference strains ^c					
1000	Tomato	French Guyana, Kourou	1966	1	I
K60	Tomato	USA, North Carolina	1953	1	I
K136	Tomato	BWI, Trinidad	1957	1	NK ^d
S225	Tomato	Peru, Lupuna	1966	1	III
S236	Tomato	Australia, Nambour	1965	1	III
K105	Tobacco	USA, Florida	1955	1	IV
S247	Tobacco	Colombia	NK	1	I

^aM = strains from Martinique and G = strains from Guadeloupe.

^bAccording to Prior and Steva (15).

^cRace and biovar typing are obtained from L. Sequeira (*unpublished*).

^dNK = not known.

among strains for reaction of the cultivars at the different temperatures.

Points corresponding to the 22 strains and the 15 temperature × cultivar combinations were plotted according to their coordinates in the plane defined by the first two factors F1 and F2 (Figs. 1 and 2). The elements (columns or rows) were not equally important. The absolute contribution of each element of a factor provided an estimate of the part played by that element in the variability in disease attributed to the factor. The most important elements are those with high absolute contributions. More than half of the estimates were explained by a plane that described 58% of the variability (or inertia) among the observations. The height of each bar within histograms (cultivar) was proportional to the total number of living plants, expressed by a percentage, in the five crops whose contribution to the axes was high (underlined in Fig. 1).

RESULTS

Variables corresponding to Floradel and Caraïbo were slightly dispersed in contrast to those for Capitan, which are concentrated in two areas (Fig. 2). In addition, the factorial values for Capitan increased in inverse ratio to temperature especially because of the proximity of the variables corresponding to the same temperatures (25 and 28 C).

Table 2. Number of symptomless plants 21 days after inoculation with strains of *Pseudomonas solanacearum* from FWI and reference strains

Strains ^b	cv. Floradel					cv. Capitan					cv. Caraïbo				
	Trial conditions ^a														
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
MT1	1 ^c	1	1	0	2	1	3	5	3	4	4	5	8	7	8
MT2	2	2	4	2	4	3	5	7	8	9	6	7	9	10	10
MT4	4	5	4	6	6	7	8	10	9	10	10	10	10	10	10
MT5	5	4	5	4	4	3	4	8	1	1	8	8	9	7	9
GT1	2	3	0	2	1	5	3	4	0	1	9	8	7	7	9
GT4	1	1	1	2	1	1	6	5	4	9	5	6	8	7	10
GT5	8	8	10	8	9	8	6	10	9	10	10	10	10	10	10
MA1	3	3	5	1	4	5	8	10	6	10	6	7	10	10	10
MA2	2	3	5	6	4	6	8	10	9	9	10	10	10	9	10
MA3	3	2	4	4	2	3	6	7	3	7	6	8	9	9	10
GA1	6	7	6	7	6	9	10	10	10	10	9	10	10	10	10
GA2	2	1	2	4	0	2	5	7	6	8	6	7	8	10	10
GA3	7	6	8	7	3	8	9	10	10	10	10	10	10	10	10
GA4	2	2	3	1	0	3	3	5	4	7	7	8	9	4	10
GA5	2	2	3	2	0	5	4	9	3	9	6	8	7	7	10
MP1	1	1	4	2	3	4	4	6	5	3	5	5	9	4	10
GP1	2	2	2	3	2	4	4	6	5	8	5	7	8	6	10
MB1	2	2	4	4	0	6	5	8	3	8	4	5	7	7	10
ME1	7	6	10	4	5	10	8	10	2	7	10	10	10	10	8
ME2	1	1	2	2	1	4	4	6	6	7	5	6	8	5	9
ME3	4	4	3	2	4	2	2	4	3	7	6	7	9	6	9
GE1	4	5	3	6	4	3	2	4	5	4	6	7	9	5	8
Reference strains															
1000	6	6	9	8	7	8	8	10	8	10	10	10	10	10	10
K60	6	6	8	9	7	8	8	10	9	10	10	10	10	10	10
K74	5	3	8	6	5	4	3	6	5	4	8	8	9	6	10
K105	6	7	10	9	10	9	9	10	10	10	10	10	10	10	10
K136	4	4	6	7	5	5	7	7	6	4	8	8	8	5	10
S225	3	3	3	3	1	3	6	8	4	4	7	7	8	8	10
S236	8	6	10	8	7	9	9	10	9	10	10	10	10	10	10
S247	9	9	9	10	9	10	10	10	10	10	10	10	10	10	10

^aTemperature conditions in growth chambers were 1 = 32 ± 2 C, 2 and 3 = 28 ± 2 C, 4 and 5 = 25 ± 2 C.

^bM = strains from Martinique and G = strains from Guadeloupe.

^cSymptomless plants of 10 inoculated plants.

Because of the high susceptibility of Floradel, factorial values of the points on the x-axis (F1) are positive and high (Fig. 2), whereas they are low and negative for the other cultivars. On F1, which explains 38.8% of the total inertia, the most important variables correspond

to Floradel. Strains in an identical position on the factorial plane F1*F2 (positive values of F1) had little influence on the mortality of the Floradel cultivar—44 to 86% of plants did not wilt. On the other hand, the strains with negative values on F1 induced a high

level of mortality in this cultivar. Only 14–22% of tomato plants survived 21 days after inoculation.

On the y-axis (F2), which explained 19% of total inertia, the most important temperature × cultivar combinations corresponded to the Caraïbo cultivar and

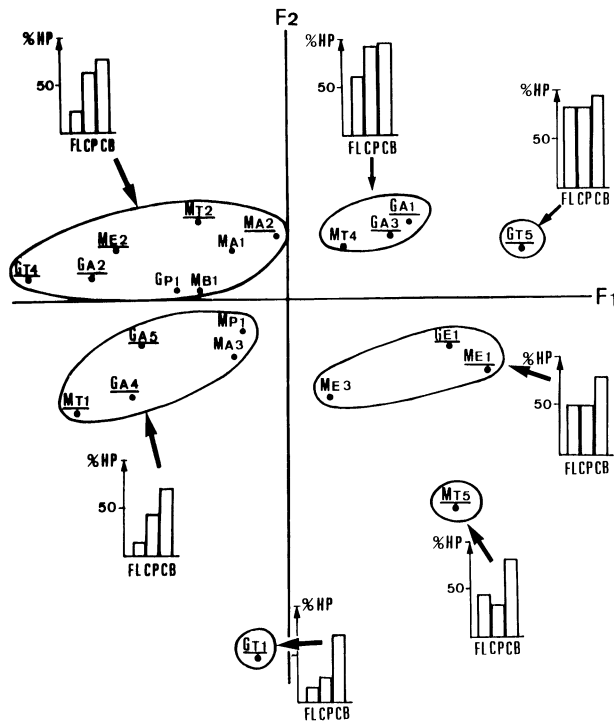


Fig. 1. Results of the correspondence analysis on data presented in Table 2. Projection of points corresponding to the different strains from FWI in the factorial plane defined with factors 1 and 2 (F1*F2). Each group of strains is characterized by a histogram of symptomless plants (%HP = percent of healthy plants) for three tomato cultivars: FL = Floradel, CP = Capitan, and CB = Caraïbo. Histograms were constructed from results achieving from distinctive strains (underlined strains) within groups (higher absolute contributions).

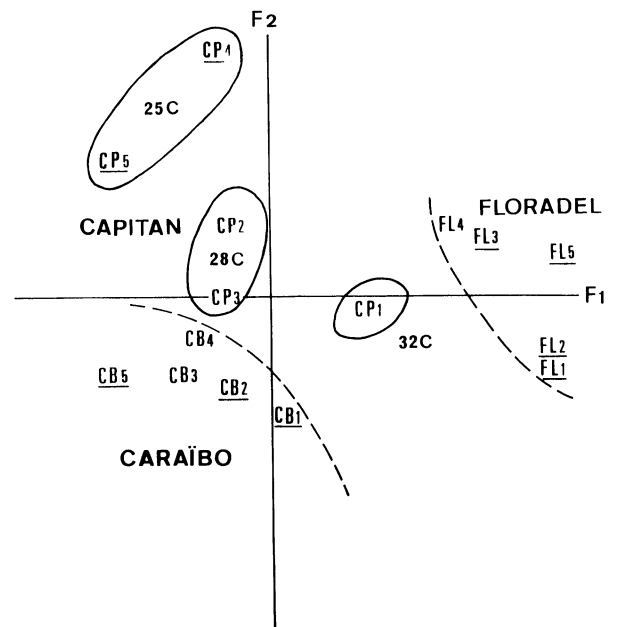


Fig. 2. Projection of points corresponding to the different variables in the factorial plane F1*F2. Letters denote tomato cultivars: FL = Floradel, CP = Capitan, and CB = Caraïbo. Numbers denote temperature conditions in five growth chambers: one set at 32 ± 2 C (I), and two each at 28 ± 2 C (2,3) and 25 ± 2 C (4,5). Relevant variables with higher absolute contributions are underlined.

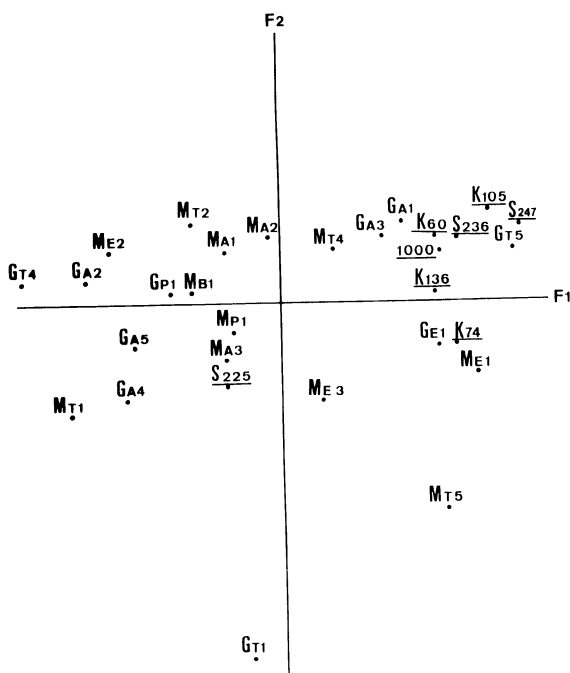


Fig. 3. Projection of reference strains as additional individuals in the factorial plane F1*F2 (Fig. 1) for comparison with strains from FWI. Reference strains are underlined.

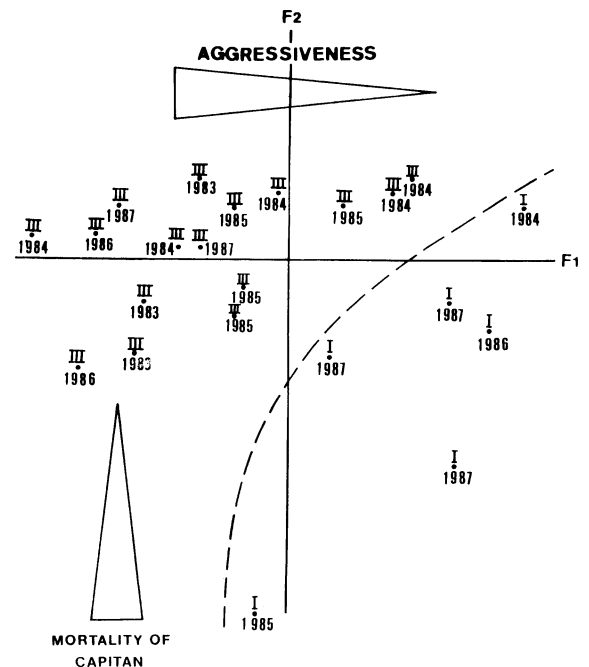


Fig. 4. Distribution of strains from FWI in the factorial plane F1*F2 with indication of year of isolation and biovar typing.

particularly to Capitan at 25 C. The strains that had little effect on Capitan (between 64 and 96% of the plants survived) had positive values on axis 2. Strains that wilted a majority of Capitan tomatoes (26 to 52% of plants survived) had values in the other half of the plane. The most aggressive strains were in the fourth quadrant, where the temperature \times cultivar combinations for Caraïbo were also found. The strains located here caused the death of the greatest number of tomato plants of the three cultivars—14, 26, and 72% of plants of Floradel, Capitan, and Caraïbo were free of symptoms, respectively.

Consequently, the strains were grouped into four different aggressiveness profiles. Strains MT4, GA1, GA3, and GT5 found in quadrant 2 caused little damage in the three cultivars. Strain GT5 was the least aggressive. In contrast, the most aggressive strains were found in the fourth quadrant. The GT1 strain stands out from this group because of its high aggressiveness on Capitan (26% compared to 44% of the plants survived). The other strains stand out in relation to their behavior with Capitan. In the first quadrant, MT2, GT4, MA2, GA2, GP1, MB1, and ME3 are less aggressive in Capitan than in Floradel, whereas in the third quadrant, ME1, ME3, GE1, and MT5 cause similar damage to these two cultivars.

All reference strains fell into the same half plane defined by the positive values of F1 with the exception of S225 (Fig. 3). Antillean strains found in the same position behaved similarly in that they caused the death of a smaller number of tomato plants of all cultivars, particularly Floradel, where 72% of plants survived 21 days after inoculation.

DISCUSSION

The inoculum concentration used in these experiments was low to avoid masking differences among strains caused by high mortality. The majority of the strains from Guadeloupe and Martinique were more aggressive than the reference strains. This may explain why cultivars selected in the Caribbean for resistance to bacterial wilt express high resistance when planted in other tropical areas (5). The strains from Guadeloupe were similar in aggressiveness to those from Martinique. Thus, the severe bacterial wilt of eggplant in Martinique that has occurred since 1983 can not be simply explained by an enhanced aggressiveness of strains located on that island.

Both Capitan and Caraïbo possessed resistance to bacterial wilt. On the factorial plane (Fig. 2), the variables corresponding to the susceptible cv.

Floradel all have high positive abscissae, which is in contrast to those of the Capitan and Caraïbo cultivars which have low negative abscissae. Resistance of Capitan is different from that of Caraïbo which was not temperature dependent, as shown by the position of the points corresponding to these two cultivars in relation to the origin of the ordinates. For Caraïbo, the variables are grouped together. In contrast, those of Capitan are highly dispersed; its resistance depends on temperature. At high temperatures (32 C), Capitan becomes as susceptible as Floradel. A loss of resistance at high temperatures has been reported for other cultivars (9,12,14,16). Therefore, Capitan probably cannot be grown in the FWI during the hot season, even though its agronomic behavior (fruitage, yield) is superior to that of Caraïbo at this period of the year (5).

Aggressiveness varied greatly among strains of *P. solanacearum* in the Antilles. The largest differences were observed after inoculation of Capitan. Resistance of this cultivar appears to be similar to Caraïbo when put in contact with strains of biovar III, which have a positive ordinate, but more like Floradel if inoculated with strains of biovar I. Thus, resistance of a cultivar can vary among different geographic locations depending on the strains of *P. solanacearum* present (11).

Strains that are relatively aggressive on Capitan belonged to biovar I (Fig. 4), whereas the other strains studied are of biovar III. A linkage between biovar and aggressiveness was not observed in results from the other cultivars. The strains of biovar III which are in the majority in the two islands may be endemic to the Antilles. In contrast, biovar I could have been introduced into the islands with imported vegetable matter of diverse geographic origin, which may explain the notable differences in aggressiveness between the different strains of this biovar. Alternatively, biovar I could be endemic but not as well adapted to the agropedoclimatic conditions in the Antilles.

Finally, no relation between the date of isolation and the aggressiveness of the strains has been identified (Fig. 4). Thus, the length of preservation would not seem to be responsible for the differences in aggressiveness observed in these tests.

Programs for improvement of tomatoes for resistance to bacterial wilt should include strains belonging to each of the groups reported here. In addition, the exclusive cultivation of Caraïbo in the Antillies may exert strong selection pressure (18) and could cause a shift in the populations of *P. solanacearum* toward types that are more aggressive to this

cultivar. The present study could be a starting point for followup work on the evolution of populations of *P. solanacearum* in these two islands.

ACKNOWLEDGMENTS

We thank M. Bérarnis and Z. Bernard for technical assistances in this study and J. A. Bartz for his kind advice during the writing of this paper.

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