Hypersensitivity in *Capsicum chacoense* to Race 1 of the Bacterial Spot Pathogen of Pepper

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ABSTRACT

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Plants of *Capsicum chacoense* PI 260435 were found to respond in a hypersensitive manner to pathotype 1 of the pepper strain of *Xanthomonas campestris* pv. *vesicatoria*. Infiltration inoculation with 10^8 bacterial cells per milliliter followed by incubation at 30 C caused visible collapse of inoculated leaf tissues within 24 hr. Leaf lesions did not enlarge beyond the areas originally infiltrated. The concentration of bacteria in inoculated leaf tissue was greatly reduced during the first 24 hr of incubation, while electrolyte loss from such tissue was increased during the same period. Evidence was obtained that hypersensitivity (resistance) from *C. chacoense* to pathotype 1 resulted from a dominant, genetic factor.

Bacterial spot of pepper (*Capsicum* sp.), caused by *Xanthomonas campestris* pv. *vesicatoria* (Doidge) Young et al (*X. vesicatoria*) (8), is difficult to control with foliar sprays under conditions of high humidity and frequent rain. Resistance in pepper was found in PI 163192 (*Capsicum annuum* L.) (7), but the existence of pathotypes of the bacterium was discovered in early field tests of breeding lines (4). One pathotype of the bacterium occurs in many locations throughout the world (5). Resistance from PI 163192 to race 2 (pepper strain) (PR2) has been characterized as a hypersensitive response (HR) (3) and distinguished from the HR in pepper caused by the tomato strain of the bacterium (1). This study was designed to characterize the response of *C. chacoense* PI 260435 (Cc 435) to inoculation with PR1 and compare this response with the hypersensitivity to PR2 found originally in *C. annuum* PI 163192.

MATERIALS AND METHODS

Inocula standardized colorimetrically to 50% light transmission (=10^8 cells/ml) were prepared with bacterial cells harvested by centrifugation of 24-hr, agitated, nutrient broth cultures. Inoculation was accomplished by hypodermic infiltration of intercostal leaf tissues (6) and inoculated plants were incubated at 30 C in temperature-controlled (±2 C) rooms.

Seed for test plants of Cc 435 was obtained from the Southern Regional Plant Introduction Station, Experiment, GA. Seeds of FLA 23-1 (23-1), a homozygous line possessing dominant (single-factor) resistance to the PR2 strain of the bacterium derived from PI 163192, were on hand. Florida VR-2 (VR-2) (2), homozygous for multiple virus resistances in addition to hypersensitive resistance (from PI 163192) to the PR2 strain of the bacterium, was used as a recurrent parent in a breeding program to transfer resistance to PR1 from *C. chacoense* to *C. annuum*. Resistant plants from each generation were backcrossed to VR-2 to obtain the next generation.

Bacterial populations in vivo were determined from replicated samples of inoculated leaf tissue harvested at various intervals after inoculation (7). Leaf tissue replicates were triturated in 0.5 ml sterile tap water, and after appropriate dilution for counting colonies later, 0.5 ml of bacterial suspension was spread on a plate of nutrient agar. The resulting colonies were counted after 3 days of incubation at 30 C. Each harvest was replicated three times.

Electrolyte loss evaluations were determined from 10.17 cm^2 inoculated leaf tissue harvested at timed intervals after inoculation and suspended in 12 ml distilled water (3). Conductivity in μmhos of the suspending water was recorded immediately and again after shaking for 1 hr at 25 C. The difference between the two readings was considered indicative of the influence of the bacteria on host tissue.

RESULTS

Bacterial populations during the first 12 hr after leaf inoculation of Cc 435 remained essentially the same (Table 1). During the next 12 hr of incubation, or when the first evidence of tissue collapse in inoculated leaves became apparent, fewer bacteria were recovered. Most completely inoculated leaves fell off after 24 hr of incubation. This pattern of in vivo bacterial population change was similar to that determined by assays from leaves of 23-1 inoculated with PR2 (7). However, the results from these two inoculations were different from those obtained using leaves of (susceptible) 23-1 infected with PR1. In this latter combination, a progressive, nearly 20-fold increase in bacterial population in vivo occurred during the first 24 hr of incubation. After 48 hr of incubation, the bacterial population remained much higher in these latter leaves than in the 23-1/PR2 and Cc 435/PR1 leaves.

Electrolyte loss from leaves of Cc 435 inoculated with PR1 followed a pattern very different from that of 23-1 leaves inoculated with PR1 (Fig. 1). Leakage from inoculated leaf tissue of Cc 435 before symptoms appeared, ie, the first 12 hr of incubation, was relatively minor. Increase in electrolyte loss occurred during the next 12 hr of incubation and was comparable to electrolyte loss from 23-1 leaves inoculated with PR2 (3). Increase in loss of electrolytes from 23-1 leaves inoculated with PR1 was more gradual but continued for 48 hr after inoculation.

Plants of Cc 435 were uniformly resistant (HR) to inoculation with PR1 (Table 2). Progeny plants (F_1) of Cc 435 crossed with VR-2 also were resistant, but segregation occurred in F_2 progenies.

Table 1. Numbers of colonies (× 10^5) recovered from pepper leaves inoculated with *Xanthomonas campestris* pv. *vesicatoria* incubated at 30 C

Pepper cultivar[a]	Bacterial pathotype[b]	Incubation time (hr)					
		0	3	6	12	24	48
Cc	1	3.1[c]	5.3	4.3	2.5	0.2	0.9
23-1	1	6.2	9.0	14.2	10.6	120.2	82.0
23-1	2	0.6	1.9	10.5	18.0	0.2	0.0

[a] Cc = *Capsicum chacoense* PI 260435; 23-1 = homozygous breeding line FLA 23-1.
[b] *X. campestris* pv. *vesicatoria* pepper strains 1 and 2.
[c] Each figure represents the average of six replicates.

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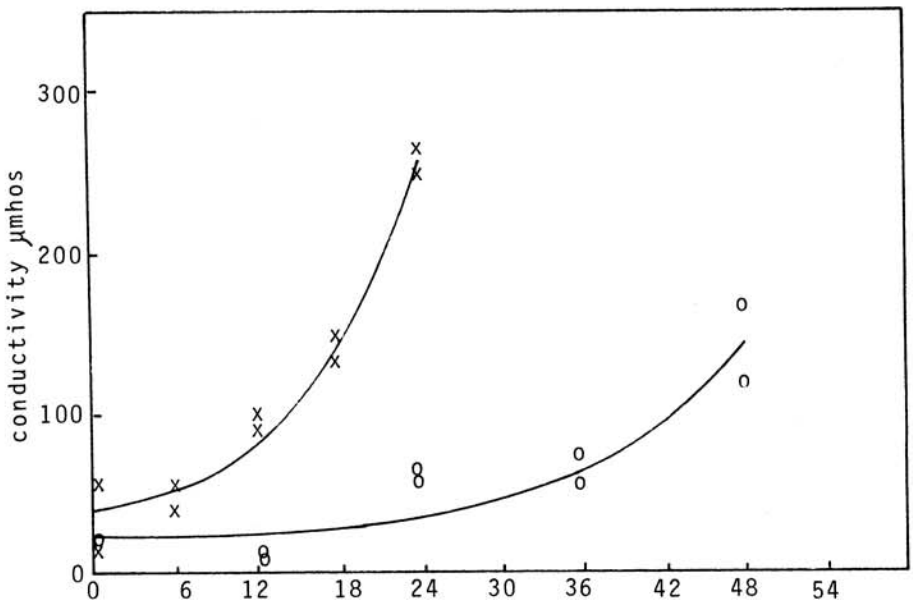

Fig. 1. Electrolyte loss from pepper leaf tissue (x = *Capsicum chacoense* PI 260435; o = *Capsicum annuum* cv. Florida 23-1) inoculated with race 1 of *Xanthomonas campestris* pv. *vesicatoria*.

Table 2. Cumulative results from inoculation of progenies of *Capsicum chacoense* PI 260435 × *Capsicum annuum* Florida VR-2 inoculated with *Xanthomonas campestris* pv. *vesicatoria* pepper strain 1

Plant or progeny[a]	Number of progenies inoculated	Number of plants	
		Resistant	Susceptible
P_r	...	35	0
P_s	...	0	38
F_1	4	43	1
$F_1BC(P_s)$	15	239	204
F_2	4	72	31

[a] P_r = resistant parent (*C. chacoense*), P_s = susceptible parent (*C. annuum*), and $F_1BC(P_s)$ = combined results from inoculation of progenies from successive generation backcrosses of resistant plants to *C. annuum*, Florida VR-2.

Within four F_2 progenies that included 103 plants, 72 plants were resistant (HR). Persistent, interspecific incompatibility and the necessity to increase fruit size (fruits of Cc 435 measured about 7 × 10 mm) as well as seed yield prompted repeated backcrossing only to *C. annuum*, ie, VR-2. The combined total of inoculated plants from 15 backcrosses (to VR-2) progenies included 239 resistant (HR) and 204 susceptible plants.

DISCUSSION

Although hypersensitive resistances for PR1 and PR2 were found in different species of *Capsicum*, responses of the two plant species were similar. As the viable population of bacteria in vivo was reduced during the first 24 hr of incubation, electrolyte loss from inoculated leaf tissue was increased. With the HR in pepper caused by the tomato strain of *X. vesicatoria*, both bacterial population in vivo and electrolyte loss increased gradually but simultaneously (1). The relation of decreasing numbers of bacteria in vivo to progressive hypersensitivity (electrolyte leakage) in infected leaf tissue remains to be determined.

Determination of inheritance of the HR derived from Cc 435 was complicated because of interspecific incompatibility of *C. chacoense* and *C. annuum*; even though the two species could be crossed with comparative ease, sterility was a major factor in F_1 plants. A few F_1 plants eventually set some fruits that contained two to five seeds each. Backcrossing was not attempted with Cc 435, and plants in successive backcross (always to VR-2) generations were progressively more fertile but decidedly less fertile than selfed *C. annuum*.

In Florida, climatic conditions are often conducive for bacterial spot development. Both pepper pathotypes occur widely within the state and either can independently cause serious loss. Resistance to only one bacterial pathotype is ineffective because the pathotype to which the planted cultivar is not resistant can exact loss equivalent to that caused by infection of a susceptible cultivar. The combination of hypersensitive resistances to both known forms of the bacterium pathogenic on pepper offers promise for improved field control of bacterial spot. The combined bacterial spot resistances have also been added to potyvirus (tobacco etch and potato Y) resistances previously available in VR-2 to provide a bell pepper type resistant to most of the serious field diseases in Florida.

LITERATURE CITED

1. Cook, A. A. 1973. Characterization of hypersensitivity in *Capsicum annuum* induced by the tomato strain of *Xanthomonas vesicatoria*. Phytopathology 63:915-917.
2. Cook, A. A., Ozaki, H. Y., Zitter, T. A., and Blasquez, C. H. 1976. Florida VR-2, a bell pepper with resistance to three virus diseases. Fla. Agric. Exp. Stn. Circ. S-242. 6 pp.
3. Cook, A. A., and Stall, R. E. 1968. Effect of *Xanthomonas vesicatoria* on loss of electrolytes from leaves of *Capsicum annuum*. Phytopathology 58:617-619.
4. Cook, A. A., and Stall, R. E. 1969. Differentiation of pathotypes among isolates of *Xanthomonas vesicatoria*. Plant Dis. Rep. 53:617-619.
5. Cook, A. A., and Stall, R. E. 1982. Distribution of races of *Xanthomonas vesicatoria* pathogenic on pepper. Plant Dis. 66:388-389.
6. Klement, Z. 1963. Rapid detection of the pathogenicity of phytopathogenic pseudomonads. Nature 188:299-300.
7. Stall, R. E., and Cook, A. A. 1966. Multiplication of *Xanthomonas vesicatoria* and lesion development in resistant and susceptible pepper. Phytopathology 56:1152-1154.
8. Young, J. M., Dye, D. W., Bradbury, J. F., Panagopoulos, C. G., and Robbs, C. F. 1978. A proposed nomenclature and classification for plant pathogenic bacteria. N.Z. J. Agric. Res. 21:155-177.