Expression of Resistance to *Xanthomonas campestris* pv. *phaseoli* in *Phaseolus vulgaris* Under Tropical Conditions

D. M. WEBSTER, Twin Falls, ID 83316, and S. R. TEMPLE and G. GALVEZ, CIAT, Cali, Colombia

ABSTRACT

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The bean (*Phaseolus vulgaris*) cultivar Jules and the PI 207262 line are moderately resistant to *Xanthomonas campestris* pv. *phaseoli* in the temperate zone but appeared susceptible in field evaluations in the tropics. Susceptibility to the pathogen was related to environmental factors that caused very early maturity and poor growth; blight resistance was expressed relative to controls when these factors were altered to permit more normal growth of Jules and PI 207262. The effect of environmental factors on resistance to common blight was observed in evaluations made at 44 and 55 days after planting but not at 34 days.

Common bacterial blight, caused by *Xanthomanas campestris* pv. *phaseoli* (Smith) Dye (XCP), is a serious disease of beans (*Phaseolus vulgaris* L.) in much of the lowland tropics. One of the goals of the bean program at the Centro Internacional de Agricultura Tropical (CIAT, Cali, Colombia) is the development of cultivars with resistance to XCP. A principal step in achieving this goal is the identification of sources of resistance to this pathogen. Two of the most promising sources of genetic resistance, according to screening carried out in the United States, are PI 207262 and certain Great Northern cultivars, such as Jules (2–4). The common bacterial blight resistance of Jules and similar sources has been studied carefully in controlled environments and is generally highly heritable and expressed in a very stable manner (5,11). Furthermore, although pathogenic variation in XCP has been demonstrated, blight resistance is generally expressed relative to susceptible controls (6,8,10). In repeated evaluations in the field in the tropics, however, Jules and PI 207262 performed as if they were susceptible to XCP (CIAT, *unpublished*).

Under the short tropical day length at CIAT, Jules and PI 207262 are induced to flower very early and produce a stunted plant compared with locally grown cultivars. In addition, Jules and PI 207262 are relatively susceptible to fungi that cause such blight diseases as Alternaria blight (*Alternaria* sp.) and angular brown leaf spot (*Isariopsis griseola* Sacc.) (= *Cercospora* sp.?), which are not severe on locally grown cultivars.

Andrus (1) has discussed the effect of plant vigor on expression of resistance in beans to bacterial pathogens. It seemed possible that the susceptible reaction of Jules and PI 207262 in the tropics did not result from inadequacy of resistance factors in the host but from poor growth of the host. The purpose of this investigation was to determine the effects of other foliar blights and day length on the expression of resistance to common bacterial blight in dry beans.

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MATERIALS AND METHODS

The XCP strain Xp123, originally isolated from dry beans at CIAT, was one of the most virulent in an evaluation of strain variability (CIAT, *unpublished*). The culture was lyophilized from a suspending medium of 5% peptone and 10% sucrose. Stock cultures were obtained by streaking lyophilized material onto yeast-dextrose carbonate agar (YDCA) (9), grown for 1 day at 28 C, then stored at 4 C for as long as 2 mo. Refrigerated stocks were not transferred again; when viability dropped, they were replaced by streaking from another lyophilized culture.

Beans were planted at CIAT in 8-m rows, 60 cm between rows, and after 10 days, seedlings were thinned to 40 cm between plants. Furrow irrigation was used when necessary. Main plots were day length and fungicide applications, and each was arranged with subplots, *Phaseolus* lines, in a split-split block design of four blocks and three replicates (7). Within each block, there was one row of each of five lines of beans: *P. acutifolius* Grey line P597 and dry bean cultivars Jules, PI 207262, Porrillo Sintetico (Porrillo), and Puebla 152, randomized separately within each block. The *P. acutifolius* line P597, which was highly resistant to XCP, was included as a check so that any mechanical damage resulting from inoculation could be recognized. Jules and PI 207262 are considered moderately resistant to XCP in the temperate zone (2,3). Both grow in Colombia as prostrate vines, although Jules has a more erect growth habit than PI 207262. Porrillo and Puebla 152 are susceptible to XCP but are otherwise well adapted to the tropics. Porrillo has an erect vine growth habit, whereas Puebla 152 grows as a prostrate vine.

Natural day length at CIAT is always close to 12 hr. Supplemental lighting provided by two 300W, 200V incandescent bulbs suspended 2 m above the ground and 2 m from the ends to the center row extended the photoperiod to 16 hr. A 15-m unplanted buffer zone separated blocks with lights from blocks without lights.

The fungicide treatment consisted of alternate spray applications of benomyl at 0.5 kg/ha and manzeb at 3 kg/ha at 2-wk intervals beginning 3 wk after planting. Benomyl is a systemic fungicide and controls Alternaria leaf spot and angular brown leaf spot as well as many other diseases caused by fungi. Manzeb controls rust caused by *Uromyces phaseoli* (Reben) Wint. var. *typica* Arth. as well as many of the same diseases controlled by benomyl. Neither fungicide is considered to have any effect on common bacterial blight. A row of the cultivar Porrillo separated blocks with fungicides from blocks without fungicides. In addition, a row of Porrillo bordered each replicate. Replicates were separated by five rows of maize (*Zea mays* L.) and sorghum (*Sorghum bicolor* (L.) Moench).

Plants were inoculated with XCP strain Xp123 at weekly intervals beginning 3 wk after planting and continuing for the duration of the experiment. Inoculum was prepared from the growth of 2-day-old plate cultures on YDCA and diluted with water to an absorbance at 600 nm that corresponds with 5×10^7 colony-forming units per milliliter. This suspension was sprayed onto plants at 3 kg/cm^2, with the sprayer nozzle held at least 3 m from plants to avoid excessive mechanical damage.

Disease evaluations were made at 34,

44, and 55 days after planting and were based on a severity index that considered lesion size and proportion of leaves infected. The entire experiment was conducted twice, during December 1975–February 1976 and April–June 1976.

Table 1. Effect of day length on induction of flowering of *Phaseolus* sp. in the field at CIAT

	Days to flower	
Line	12-Hr day length	16-Hr day length[a]
P597[b]	35	45
Jules	29	45
PI 207262	30	43
Puebla 152	36	43
Porrillo	40	47

[a] Day length extended 4 hr with light from incandescent bulbs.
[b] *P. acutifolius*; other four lines are *P. vulgaris*.

Table 2. Analysis of variance of effects of day length (D) and fungicide (F) applications on severity of common bacterial blight on *Phaseolus* sp. lines (L)

Days after planting	Source[x]	df	Mean square[y]
34	L	3	14.0502**
	L × D	3	0.0502
	L × F	3	0.0368
	L × D × F	3	0.0203
	E_d[z]	24	0.0253
44	L	3	6.3915**
	L × D	3	1.6666**
	L × F	3	0.0700
	L × D × F	3	0.0430
	E_d	24	0.0518
55	D	1	7.6400*
	E_a	2	0.1722
	F	1	0.7931*
	E_b	2	0.0368
	L	...	3.4809**
	L × D	...	1.7132**
	L × F	...	0.2292*
	L × D × F	...	0.0426
	E_d	...	0.0684

[x] Only sources of treatment effects that are significant, and interactions with lines, are listed.
[y] * = $P<0.05$; ** = $P<0.01$.
[z] E = error.

RESULTS

An analysis of variance was not made on data from the first experiment, December 1975–February 1976, because severe damping-off led to low plant populations. The days to flower and disease reactions of both experiments were very similar; therefore, only the results of the second experiment are reported. Data from the P597 line were omitted from the analysis because P597 remained symptomless and the variance associated with it was zero.

The flowering date for each line was recorded when 50% of the plants had produced one or more flowers. All five lines were day length sensitive, but the difference in flowering date between 12- and 16-hr day lengths was greatest for Jules and PI 207262 (Table 1). With the 16-hr day length, there was a range in the flowering dates among lines of only 4 days.

At 34 days after planting, Jules was the most resistant of the *P. vulgaris* lines and Puebla 152 and Porrillo were the most susceptible to XCP. At this first evaluation date, there was no effect of day length or fungicide on disease severity ratings (Tables 2 and 3).

By 44 days after planting, there was a great effect of day length on disease severity of Jules and a smaller effect (not significant, $P>0.05$) on disease severity of PI 207262. Day length did not have any apparent effect on disease severity of Puebla 152 or Porrillo (Table 3). This differential response of lines to day length gave a significant ($P<0.01$) interaction (L × D) in the analysis of variance (Table 2).

Data from the final evaluation date at 55 days showed a great effect of day length on disease severity of Jules and PI 207262 (Table 3). Disease ratings for Puebla 152 and Porrillo also tended to be less severe with long days but not significantly so ($P>0.05$) (Tables 2 and 3).

The analysis of data from the evaluation at 55 days also showed a significant ($P<0.05$) interaction of fungicides with lines. The source of this interaction was an effect of fungicides on common bacterial blight severity of PI 207262, and to a lesser extent, on that of Jules. There was no effect on disease severity of the two adapted lines, Porrillo and Puebla 152.

DISCUSSION

The differential effect of fungicides indicates a greater susceptibility of PI 207262, and to a lesser extent, of Jules to fungi as opposed to XCP. The fungi that cause angular brown leaf spot and Alternaria blight often infect tissue that is under stress or is senescent, and under these conditions, these fungal diseases cause symptoms that resemble those of common bacterial blight. It is also possible that secondary infections by weakly pathogenic fungi could increase the severity of common bacterial blight. Benomyl and manzeb had no effect on disease severity of the inoculated adapted lines, Porrillo and Puebla 152.

Coyne et al (5) studied the effects of maturity on disease severity and on populations of bacteria in leaves of bean lines resistant and susceptible to XCP and concluded that susceptibility increases as plants mature. Webster et al (11) evaluated the resistance to XCP of greenhouse-grown seedlings and adult plants of F_4 families from crosses involving Jules and PI 207262 as parents. In the greenhouse, there was little difference among families in rates of maturity and there was only a small effect of maturity on family rank; resistance to the pathogen may have declined, but it was still expressed, relative to blight susceptible families, even after pod set.

In this study, maturity of Jules and PI 207262 was influenced by day length, and disease severity in the second and third

Table 3. Effects of day length, fungicide application, and time of evaluation on severity of common bacterial blight on five *Phaseolus* sp. lines

Days after planting	Day length (hr)	Fungicide treatment[a]	Jules	PI 207262	Porillo	Puebla 152	P597	LSD$_{0.05}$ at: $D_1F_1L_1$-$D_1F_1L_2$	$D_1F_1L_1$-$D_1F_2L_1$	$D_1F_1L_1$-$D_2F_1L_1$
34	16	Yes	1.35[b]	3.38	3.70	3.65	1.00	0.27[c]	0.30	0.69
		No	1.35	3.07	3.69	3.70	1.00			
	12	Yes	1.40	3.09	3.81	3.70	1.00			
		No	1.57	3.04	3.76	3.67	1.00			
44	16	Yes	1.65	3.11	3.93	3.96	1.00	0.38	0.39	0.76
		No	1.61	3.25	3.99	3.97	1.00			
	12	Yes	3.16	3.50	4.00	4.08	1.00			
		No	3.27	3.75	3.72	4.07	1.00			
55	16	Yes	2.35	3.19	4.32	4.08	1.00	0.44	0.48	0.77
		No	2.42	3.91	4.44	4.30	1.00			
	12	Yes	4.01	4.19	4.71	4.33	1.00			
		No	4.37	4.78	4.74	4.27	1.00			

[a] Alternately applied benomyl at 0.5 kg/ha and manzeb at 3 kg/ha at 2-wk intervals from April to June 1976.
[b] Average disease severity. Severity index: 1 = no visible lesions, 2 = lesions covering less than 25% of the leaf surface on fewer than 10% of the leaves, 3 = lesions covering less than 25% of the leaf surface on 10–50% of the leaves, 4 = lesions covering more than 25% of the leaf surface on 10–50% of the leaves, and 5 = lesions covering more than 25% of the leaf surface on more than 50% of the leaves.
[c] Least significant difference for lines (L), fungicides (F), and day length (D): D, 12 or 16 hr; F, yes or no; L, between any of the five *Phaseolus* sp. lines.

evaluations indicated that this influence was important to the expression of common bacterial blight resistance by Jules and PI 207262. When grown under natural day length (12 hr), Jules and PI 207262 developed small plants that were not vigorous and that became senescent relatively early. Relative to susceptible but adapted lines, common bacterial blight resistance of Jules began to decline after the first evaluation.

When grown under long day conditions PI 207262 and Jules matured at a rate similar to that of the other lines. Although disease severity increased on all the *P. vulgaris* lines during the observation period, Jules expressed a useful level of resistance to XCP, compared with the others, through the third evaluation, when all plants were bearing pods.

The line PI 207262 appeared to be less resistant than Jules to XCP in this study and also in greenhouse studies involving the strain Xp123 (11). Although Xp123 did not represent all the variation possible within XCP, it did serve to demonstrate the possible effects of environment on expression of common bacterial blight resistance. PI 207262 is reported to be relatively more resistant than Jules to other XCP strains (8,10).

Differences in plant growth and physiology in the first evaluation were apparently insufficient to affect disease severity. When evaluating diverse germ plasm in the tropics under normal field conditions, it seems essential to develop a high incidence of disease early in the growing season and evaluate for resistance to XCP then because later in the season problems with adaptation can confuse results. Common bacterial blight resistance should then be verified at different growth stages, possibly in controlled environments, to ensure that the resistance is effective in mature plants (4).

Greenhouse and field evaluations of common bacterial blight resistance do not always agree. In this example, the discrepancy resulted from specific environmental conditions that caused poor growth of lines otherwise resistant to the pathogen rather than from intensity of disease under field conditions. Jules and PI 207262 have proved useful in the CIAT breeding program as factors conditioning resistance to XCP are tranferred to adapted genotypes (*unpublished*).

LITERATURE CITED

1. Andrus, C. F. 1948. A method of testing beans for resistance to bacterial blights. Phytopathology 38:757-759.
2. Coyne, D. P., and Schuster, M. L. 1970. 'Jules,' a Great Northern dry bean variety tolerant to common blight bacterium (*Xanthomonas phaseoli*). Plant Dis. Rep. 54:557-559.
3. Coyne, D. P., and Schuster, M. L. 1973. *Phaseolus* germplasm tolerant to common blight bacterium *Xanthomonas phaseoli*. Plant Dis. Rep. 57:111-114.
4. Coyne, D. P., and Schuster, M. L. 1974. Inheritance and linkage relations of reaction to *Xanthomonas phaseoli* (Smith) Dowson (common blight), stages of plant development and plant habit in *Phaseolus vulgaris* L. Euphytica 23:195-204.
5. Coyne, D. P., Schuster, M. L., and Hill, K. 1973. Genetic control of reaction to common blight bacterium in bean (*Phaseolus vulgaris*) as influenced by plant age and bacterial multiplication. J. Am. Soc. Hortic. Sci. 98:94-99.
6. Ekpo, E. J., and Saettler, A. W. 1976. Pathogenic variation in *Xanthomonas phaseoli* and *X. phaseoli* var. *fuscans*. Plant Dis. Rep. 60:80-83.
7. Little, T. M., and Hills, F. J. 1972. Statistical Methods in Agricultural Research. University of California Agricultural Extension, Berkeley. 242 pp.
8. Schuster, M. L., Coyne, D. P., and Hoff, B. 1973. Comparative virulence of *Xanthomonas phaseoli* strains from Uganda, Colombia, and Nebraska. Plant Dis. Rep. 57:74-75.
9. Starr, M. P. 1944. Studies of phytopathogenic bacteria. Ph.D. dissertation, Cornell University. (Diss. Abstr. 1944:30).
10. Valladares-Sanchez, N. E., Coyne, D. P., and Schuster, M. L. 1979. Differential reaction of leaves and pods of *Phaseolus* germplasm to strains of *Xanthomonas phaseoli* and transgressive segregation for tolerance from crosses of susceptible germplasm. J. Am. Soc. Hortic. Sci. 104:648-654.
11. Webster, D. M., Temple, S. R., and Schwartz, H. F. 1980. Selection for resistance to *Xanthomonas phaseoli* in dry beans. Crop Sci. 20:519-522.