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## Sources of Resistance to Common Bacterial Blight and Rust in Elite *Phaseolus vulgaris* L. Germplasm

E. Arnaud-Santana<sup>1</sup>, M.T. Mmbaga<sup>2</sup>, D.P. Coyne<sup>3</sup>, and J.R. Steadman<sup>4</sup> *University of Nebraska, Lincoln, NE 68583-0724* 

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Abstract. We studied leaf and pod reactions of 18 Phaseolus vulgaris germplasm lines (three temperate and 15 tropical) to four Xanthomonas campestris pv. phaseoli (XCP) (Smith) Dye strains and seven Uromyces appendiculatus (UA) (Pers.) Unger races. Line  $\times$  XCP interaction was significant for leaf and pod reactions. The common bean lines XAN-159, BAC-6, and XAN-112 had the best combined leaf and pod resistance to XCP. Line  $\times$  UA race interactions were significant (P = 0.05). Lines IAPAR-14 and BAC-6 had the best combined resistance to XCP and UA.

Common bacterial blight incited by *Xanthomonas campestris* pv. *phaseoli* (XCP), and rust caused by *Uromyces appendiculatus* (UA) are limiting constraints in the commercial production of dry beans worldwide (Sanders and Schwartz, 1980). Management strategies available for blight control involve specific cultural practices that can compromise a grower's crop management flexibility. Currently, fungicide use is the main control strategy for rust-susceptible cultivars (Steadman and Lindgren; 1983). The cost of fungicides makes it difficult for many growers, especially small landholders, to produce beans profitably. Thus, using dry bean cultivars resistant

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or tolerant to these pathogens is recommended as the most cost-effective management method (Sanders and Schwartz, 1980). Disease-resistant cultivars permit profitable commercial production of dry beans in regions where rust, common blight, or both reduce yields. Germplasm resistant to XCP has been identified (Coyne and Schuster, 1969, 1983; Mohan, 1981) [S. Beebe, Centro Internacional de Agricultura Tropical (CIAT) Cali, Colombia, personal communication], but most sources have not been tested for leaf and pod reaction using diverse XCP strains from temperate and tropical origins. In addition, only a few sources of resistance to XCP in beans are known (Schuster et al., 1983). Rust and common blight resistance are usually needed in breeding programs, and the germplasm should be identified for its reaction to both pathogens.

Objectives of this research were to test known common bacterial blight-resistant *Phaseolus vulgaris* germplasm of diverse origins for leaf and pod reactions to four XCP strains and seven rust races from tropical and temperate origins.

The origin, source, and published reaction of the *P. vulgaris* lines to XCP used in this study are listed in Table 1. The seven lines received from S. Beebe were the most resistant of the 18,000 plant introductions (PIs) tested at CIAT through Feb. 1989. Seeds were sown in 15-cm (1.8-liter) clay pots, two seeds per pot, in a medium of 1 sand : 1 peat : 1 vermiculite: 1 silty clay loam (by volume) and maintained in a greenhouse at a  $25/22 \pm 2C$ 

Table 1.Origin and reaction of *Phaseolus vulgaris* germplasm to *Xanthomonas campestris* pv. *phaseoli* (XCP).

(ACI).				
Cultivar or line	Origin <sup>z</sup>	Reaction to XCP	Source	
GN Neb. no. 1 sel. 27	UNL	Resistant	Coyne and Schuster (1983	
GN Tara	UNL	Resistant	Coynne and Schuster (1983)	
XAN-159	CIAT	Resistant	McElroy, CIAT (1985)	
XAN-112	CIAT	Resistant	Annual Report (1984)	
XAN-91	CIAT	Resistant	Annual Report (1983)	
PI 207262	Colombia	Resistant	Coyne and Schuster (1983)	
BAC-5	IAPAR	Resistant	Mohan (1981)	
BAC-6	IAPAR	Resistant	Mohan (1981)	
IAPAR-14	Goiana, Brazil	Resistant	M.D. Thung (1989)	
IAPAR-16	Goiana, Brazil	Resistant	M.D. Thong (1989)	
Tamaulipa 9-B (G 04399)	CIAT	Resistant	S. Beebe <sup>y</sup>	
MSU 183 (G 06700)	CIAT	Resistant	S. Beebe	
Calima 9 (G 06772)	CIAT	Resistant	S. Beebe	
PI 209.481 (G 16836)	CIAT	Moderately resistant	S. Beebe	
RKN <sup>x</sup> (G 18443)	CIAT	Moderately resistant	S. Beebe	
ODCSJ <sup>w</sup> (G 18168)	CIAT	Moderately resistant	S. Beebe	
G 19195A	CIAT	Moderately resistant	S. Beebe	
PC-50	Dominican Republic	Susceptible	Schuster et al. (1983)	

UNL = Univ. of Nebraska, Lincoln; CIAT = Centro Internacional de Agricultara Tropical, Cab, Colombia IAPAR = Sundacao Instituto Agronomic do Parsma, Goiana, Brazil.

<sup>&</sup>lt;sup>2</sup>Research Associate, Dept. of Plant Pathology. <sup>3</sup>Professor, Dept. of Horticulture.

<sup>&</sup>lt;sup>4</sup>Professor, Dept. of Plant Pathology.

<sup>&#</sup>x27;Personal communication.

<sup>\*</sup>Red kidney noailles.

<sup>&</sup>quot;Ojo de Cabra, S.J. del Rio.

day/night cycle and a 12-h photoperiod. Two experiments (A and B) were conducted, each consisting of four pots replicated twice for each host. Two experiments were conducted to determine if similar reactions would be obtained with the reactions of the germplasm with the same XCP strains because of the significant effects of environment on the reaction and to include three additional rust isolates in the second experiment. Repeatability of rust reactions on the same lines with the same isolates is well known. A factorial arrangement with a randomized complete block design with two replications was used to accommodate inoculation with four XCP strains (common to each experiment) and seven UA races (one common and three different in each experiment).

Virulent XCP strains used were  $V_sS_s$ , (formerly EK- 11, Nebraska); LB-2 (Nebraska);  $V_sS_1$  (formerly DR-12, Dominican Republic); and T-37 (Puerto Rico). These strains are highly representative since not much variation has been detected from different geographical areas (unpublished data). Cultures were grown on the semi-selective medium for *Xanthamonas phaseoli* (Claflin et al., 1987) for 24 to 48 h at 26C. Inoculum was made in 0.01 M potassium phosphate buffer, pH 7.1, to a concentration of  $10^7$  colony-forming units/ml. Buffered saline was used as the control treatment.

The multiple-needle inoculation method (Andrus, 1948; Valladares-Sanchez et al., 1979) was used to inoculate the third fully expanded trifoliolate leaf of each plant. Two pods per plant at  $\approx 50\%$  pod filling also were inoculated by using a modified multiple-needle technique consisting of a 7-  $\times$  2.5-cm florist ribbon shredder holding one row of thirty-five 20-gauge  $\times$  1.5-cm needles (the same number and type of needles, held by the circular florist frog, as were used to inoculate the leaves).

Each pod in turn was punctured with a shredder along one side wall and dipped into the inoculum suspension for 5 to 10 sec.

The disease reaction on the leaves was recorded 15 days after inoculation. A 1 to 5 leaf reaction rating scale was based on necrosis, water soaking, and yellowed area on the inoculated leaf 1 is no disease symptoms, 2 is 1% to 5%; 3 is >5% to 25%; 4 is >25% to 50%; and 5 is  $\geq$  51% of the inoculated area with symptoms. Disease reactions on the pods were recorded 18 days after inoculation by measuring the sizes of the largest and smallest necrotic spots in the inoculated area and calculating a mean size for each pod. The pod reaction rating scale was as follows: 0 > 1 mm (resistant);  $1 \le 2$  mm (moderately resistant);  $2 \le 3$ mm (moderately susceptible);  $3 \le 4$  mm (susceptible);  $\geq 4$  mm (highly susceptible).

Analyses were conducted using the Statistical Analysis System (SAS, 1982). The simple linear relationship between leaf and pod scores for cultivars and lines on a per-plot basis in each experiment also was tested.

Rust cultures were selected from a collection of >200 cultures based on virulence patterns on different cultivars (Stavely et al., 1983). These isolates were chosen from a cluster analysis to represent diverse reactions (M.T.M. and J.R. S., unpublished data). The rust cultures from Honduras showed about the same virulence pattern as the races from Puerto Rico and the Dominican Republic, but differed from those from Nebraska. Three Honduras races (H87ZA3 no. 15, H87HP2 no. 19, and H87DA3 no. 17) and a Nebraska race (US86NP10no. 1) were used in Expt. A. Three other Honduras races (H87ZA no. 16, H87EAP1 no. 20, and H87QH3 no. 15) and race US86NP10 no. 1 were used in Expt. B.

The primary leaves were inoculated with UA 6 to 7 days after emergence, when they

were 35% to 65% expanded. Those same plants were inoculated with XCP when the third trifoliolate was fully expanded. No antagonistic or synergistic interaction between the two pathogens was evident visually when separate leaves were inoculated (Finke et al., 1986). Inoculum, consisting of 5 mg fresh urediniospotes suspended in 100 ml water and 40 pg Tween-20 surfactant/liter, was deposited on the abaxial surface as uniformly as possible using a hand sprayer. The leaf surface was allowed to dry before the plants were incubated in a large mist chamber for 16 h at 100%. relative humidity at 19  $\pm$  IC. Disease reaction rating was recorded 14 days after inoculation and was based on the size of uredinia on a scale of 1 to 6 (Stavely et al., 1983). The uredinia size ratings were converted to diameter measurements (in micrometers) for data analysis. A hypersensitive necrotic leaf reaction with no sportdating uredinia was considered highly resistant. Plants with a uredinia diameter <300 pm were resistant those 300 to 450 μm (but none >500 pm) were moderately resistant; those 450 to 550 µm (but none >600 µm) were moderately susceptible; and those >550 µm were susceptible (Mmbaga and Stavely, 1989).

Analyses of variance of the data were conducted using the SAS program.

Values for r were 0.59 and 0.43 for the association between the leaf and pod reactions in lines in both experiments, respectively. Significant ( $P \le 0.05$ ) germplasm host  $\times$  XCP strain interactions were found for leaf and pod reactions in both experiments. Only the mean ratings for the disease reactions to XCP in Expt. A are shown (Table 2), since they were similar to those in Expt. B.

Leaves of lines XAN-159, BAC-6, IAPAR-14, G 06700, XAN-112, IAPAR-16, and BAC-5 were resistant pods of the frost two

Table 2. Leaf and pod reactions of *Phaseolus vulgaris* germplasm accessions to strains of *Xanthomonas campestris* pv. *phaseoli* (XCP) (Expt. A, greenhouse, Lincoln, Neb., 1989-90).

Line	XCP strain									
	$V_4S_1$		$V_3S_8$		LB-2		T-37		Control	
	Leaf <sup>z</sup>	Pod <sup>y</sup>	Leaf	Pod	Leaf	Pod	Leaf	Pod	Leaf	Pod
GN Neb. no.										
1 sel. 27	3.5	2.2	2.0	0.2	2.5	0.0	3.0	3.9	0	0
GN Tara	4.5	1.8	3.0	1.7	2.5	0.0	3.5	3.6	0	0
XAN-159	2.0	0.0	2.0	0.0	2.0	0.0	1.8	0.0	0	0
XAN-112	3.2	2.8	1.0	0.2	1.3	0.3	2.5	1.9	0	0
XAN-91	2.0	2.0	3.0	3.5	3.0	5.1	1.5	1.8	0	0
PI 207262	4.0	3.7	4.0	3.0	4.0	3.1	2.8	3.3	0	0
A C - 5	2.5	1.8	1.0	0.5	3.0	0.5	3.0	3.4	0	0
BAC-6	2.7	0.3	1.5	0.0	2.0	0.2	2.8	0.6	0	0
IAPAR-14	1.0	2.1	1.0	1.0	2.0	1.5	1.0	1.3	0	0
IAPAR-16	3.0	1.4	1.8	0.0	1.8	0.0	2.5	3.9	0	0
G 04399	3.5	1.9	3.0	1.7	4.5	5.1	3.5	3.2	0	0
G 06700	2.3	1.7	1.3	0.5	2.0	0.6	2.5	2.6	0	0
G 06772	3.3	2.0	3.8	3.4	3.8	3.8	2.5	2.9	0	0
G 16836	3.0	2.7	2.5	3.9	2.8	4.6	2.0	2.6	0	0
G 18443	4.0	$NF^{x}$	3.5	NF	4.3	NF	2.5	NF	0	0
G 18168	5.0	2.8	3.5	2.5	4.0	3.0	3.3	2.4	0	0
G 19195A	4.5	3.9	4.5	4.1	4.0	3.8	'1.8	2.6	0	0
PC-50	4.5	5.0	4.5	3.9	5.0	5.2	3.5	3.6	0	0
LSD (5%)	0.9	2.3								

Leaf disease rating scale: 1 is no symptoms (highly resistant); 2 is 1% to 5%; 3 is >5% to 25%; 4 is >25% to 50%; and 5 is  $\geq$  51% of necrosis or yellowing (or both) in the inoculated area.

Pod reactions: 0<1 mm (resistant);  $1 \le 2$  mm (moderately resistant);  $2 \le 3$  mm (moderately susceptible);  $3 \le 4$  mm (susceptible); and  $\le 4$  mm (highly susceptible).  $^{5}NF = No$  flowering under long photoperiod.

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lines were the most resistant to the four bacterial strains (Table 2). IAPAR- 14, IAPAR- 16, G 06700, XAN-112, and BAC-5 pods were resistant to three of the four bacterial strains. The leaves and pods of XAN-91, GN Neb. no. 1 sel. 27, and GN 'Tara' were resistant to three of the four bacterial strains. PC-50 and PI 207262 leaves and pods were susceptible to all four strains.

Germplasm  $\times$  rust race interactions were significant (  $P \le 0.05$ ) in both experiments. Line IAPAR- 14 exhibited the broadest range of rust resistance in both experiments, being immune, resistant, or moderately resistant to six rust races, but susceptible to race H87ZA3 no. 16 (Table 3). BAC-6 was resistant to four races and moderately susceptible to the other three. PC-50 was resistant to five races, but was susceptible to two. GN Neb. no. 1 sel .27 and BAC-5 were susceptible to all races tested, while the other lines ranged from immune to moderately susceptible.

The intermediate, positive association detected for bean leaf and pod reaction to XCP strains supports previous reports on tepary bean (P. acutifolius A. Gray) (Zaiter and Coyne, 1989) and common bean (Rava et al., 1987) but disagrees with others (Coyne and Schuster, 1974, 1983; Valladares-Sanchez et al., 1979). The latter reported a lack of association between pod and leaf reactions in segregating progeny in common beans. Differential leaf and pod reactions of GN Neb. no. 1 sel. 27, XAN-91, and GN 'Tara' to the XCP strains were detected in our study. In addition, the overall positive association indicates that lines with leaf and pod resistance to XCP strains can be identified and developed by breeding.

The host genotype × bacterial strain interaction for leaves and pods agrees with that reported for tepary bean (Zaiter and Coyne, 1989) and common bean (Aggour et al., 1989),

but not with the results reported for common bean by CIAT (CIAT, 1978). Using different bean lines, bacterial strains, and environmental conditions could be the basis for the conflicting results.

Although lines G 06700, G 16836, G 18168, G 04399, G 06772, G 18443, and G 19195A were resistant or moderately resistant to XCP (S. Beebe, personal communication), only G 06700 leaves were resistant to all XCP strains used in our study, although its pods were not resistant. GN 'Tara' leaf resistance to V<sub>3</sub>S<sub>8</sub> and LB-2 was reported previously (Aggour et al., 1989). This line was susceptible to XCP strain T-37 on leaves and pods and to V<sub>4</sub>S<sub>4</sub> on leaves. PC-50 previously was susceptible to certain bacterial isolates (Schuster et al., 1983), and PI 207262 previously was resistant to other XCP isolates (Valladares-Sanchez et al., 1979). These results emphasize the need to test germplasm with several XCP strains to identify broad resistance required commercial production.

Common bean lines XAN-159, BAC-6, and XAN-112 exhibited the best combination of leaf and pod resistance to the bacterial strains tested. The resistance of these lines to other XCP isolates was reported previously (Aggour et al., 1989; CIAT, 1984). Lines IAPAR-14 and G 06700 also had a good combined leaf and pod resistance to the XCP strains used in this study.

Lines IAPAR-14 and BAC-6 had the best combined resistance to the two pathogens and would be useful to bean breeders where these XCP strains and rust races occur. These Brazilian lines had a higher level of resistance to XCP than the rust-susceptible GN Neb. no. 1 sel. 27, which has been used widely in breeding for resistance to XCP in beans worldwide and has contributed to germplasm used in developing the above Brazilian lines.

Table 3. Mean uredinia diameters (in micrometers) of primary leaves of Phaseolus vulgaris germplasm to cultures of Uromyces appendiculatus (Expts. A and B, greenhouse, Lincoln, Neb., 1989–90).

	Rust culture <sup>z</sup>							
	Expt. A				Expt. B			
Line	1	2	3	4	5	2	6	7
		Mean u	redinia dia	meter (pm	1)			
GN Neb. no. 1 sel. 27	550	717	533	558	600	700	583	550
GN Tara	592	408	425	433	575	367	617	308
XAN-159	500	575	492	517	525	558	358	542
XAN-112	525	600	583	600	517	600	450	567
XAN-91	567	542	342	525	542	600	600	$000_{h}$
PI 207262	533	592	550	533	508	625	492	542
BAC-5	567	575	558	550	575	650	575	558
BAC-6	542	300	300	300	500	300	450	300
IAPAR-14	300	325	300	300	550	300	000	300
IAPAR-16	533	300	367	300	633	300	500	350
G 04399	533	383	525	000	474		000	342
G 06700	517	325	383	342	550	342	458	358
G 06772	542	537	550	350	542	683	000	300
G 16836	483	300	509	298	550	283	483	567
G 18443	492	517	458	567	542	692	458	525
G 18168	525	522	450	333	575	558	408	408
G 19195A	300	458	375	575	500	588	375	325
PC-50	450	533	383	542	300	575	458	442
LSD <sub>0.05</sub> with expt.		3	30			3	8	

Rust culture: 1 =H872A3 no. 15; 2= US86NP10 no. 1; 3 =H87DA3 no. 17; 4= H87HP2no. 19; 5 =H87ZA3 no. 16; 6 = H87Q3 no. 15; 7 = H87EAP no. 20 (numbers 1 to 7 do not refer to published race numbers). '000 = Hypersensitive necrotic reaction with no sporulating uredinia.

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