REGISTRATION OF FIVE COMMON BEAN GERMPLASM LINES RESISTANT TO COMMON BACTERIAL BLIGHT: W-BB-11, W-BB-20-1, W-BB-35, W-BB-52, AND W-BB-11-56

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Common bacterial blight is one of the major diseases of dry beans worldwide. The disease is caused by the bacterium *Xanthomonas campestris* pv. *phaseoli (Xcp)=X. axonopodis pv. phaseoli.* Few commercial common bean (*Phaseolus vulgaris* L.) cultivars are resistant to common bacterial blight caused by Xcp. Nebraska Great Northern No. 1, Sel. 27 has been used as a standard and is recognized as having a useful level of resistance to the bacterium. However, the resistance in this cultivar is not as great as that found on some tepary beans (*P. acutifolius* Gray), (McElroy, 1985, Freytag, 1989, Zapata, 1989).

The University of Puerto Rico, the Agricultural Research Service, U.S. Department of Agriculture, and Cornell University cooperatively announced five common bean germplasm lines as: Wilkinson (W) – Bacterial Blight (BB) -11, -20-1, -35, -52, and -11-56 in 1990. These germplasm lines represent the culmination of more than 20 years of crossing and testing by Dr. R.E. Wilkinson at Cornell to pyramid mostly minor gene effects for common bacterial blight resistance caused by Xcp in the common bean (*P. vulgaris*) and nearly 10 years collaboration involving field testing, inoculation and selection of these lines in Puerto Rico (PR). This cooperative work was supported in part by grants from the U.S. Agency for International Development (AID/CM/TA-C-73-26), AID/TA-C- 1296, AID/DSAN/XII-G-0261 and CBA-UPR-18 (83-CRSP-2-2160), and the NY State Dry Bean Growers and Shippers fund.

Bacterial blight resistance is available in three lines with determinate growth habit which have been more susceptible than indeterminate beans to common blight and two indeterminate bushy vine types. Superior level of bacterial blight resistance were developed in lines W-BB-20-1 and W-BB-11-56 a bush and bushy vine type, respectively. The germplasm lines W-BB-20-1 and W-BB-11-56 (Zapata et al., 1991) have been used successfully as parental lines for breeding for resistance to common bacterial blight on recently released lines USNA-CBB-1 and USNA-CBB-2, respectively (Miklas et al., 2001a and 2001b). The other three lines are two bush or determinate types and one bushy vine or indeterminate type with a superior level of resistance to bacterial blight better than Great Northern No.1, Sel. 27. Another achievement was the development of a germplasm line with bush type and snap bean characteristics such as W-BB-52 with resistance to most of the bacteria tested.

The high level of Xcp resistance in the W-BB lines was developed by pyramiding minor genes for resistance from several sources such as GN-1, Sel.27, PI 207262, PI 180745, PI 180746, and 65859 (Table 1) primarily through a reciprocal backcross program. The reciprocal backcross process of pyramiding genes for resistance began with crossing two plants that derive resistance from two different sources. After two generations of screening the F_3 and F_4 for bacterial blight, the resistant selections were backcrossed to each parent and the resulting progenies were ultimately intercrossed. Typically one or more generations of selfing and screening for resistance followed this last cross. The progeny from the two backcrosses was screened for resistance for two generations. Additional minor genes from other sources of Xcp resistance were combined through a parallel procedure. Then these two small pyramids were combined through a reciprocal backcross procedure. It was assumed that the progeny from the

reciprocal backcross carried most of the genes for resistance from each original source. This assumption was taken because a high degree of recessiveness in the genes for resistance from many sources was observed. The reciprocal backcross procedure has the advantage of producing a higher percentage of homozygosity in which the recessive genes for resistance can be expressed. Also, it is suspected that some minor genes may be best expressed in the presence of certain other minor genes. If this is the case, it adds importance to the need for recovering "all" the resistance genes from both parents. Regardless of the real reason for why it works, experience has shown that the reciprocal backcross procedure is essential to accomplishing a satisfactory pyramiding of recessive genes for resistance.

Resistance to common blight was determined by screening promising families for their latent period (length of time between inoculation and symptom expression). Indicator plants used were 3M-152 (highly susceptible Puerto Rico line), Redkote (susceptible), and Sel 27.(resistant). The indicator plants developed symptoms at 3, 4, and 7 days after inoculation, respectively. The experiments were conducted in growth chambers at 85F. In the beginning it was possible to conduct a test every week but as soon as the incubation time increased the observation period also increased. Generally the material that was used to plant one experiment in the growth chamber consisted of one or more related groups or families together with various support material. Also, when F2 populations were screened, one or both parents were included, especially when no obvious phenotypic markers were involved, to help confirm that a cross had been obtained, as to get a measure of the effect of pyramiding genes for resistance. Plants that were saved from a screening test were transplanted to larger pots in the greenhouse and held for seed production and possible crossing. All crosses were made at Cornell with resistance screening conducted by multineedle wound inoculation (Zapata et al., 1985) of primary leaves on 8-day old seedlings with 10^8/CFU in a controlled growth chamber at about 85 F (29 C) (Zapata et al., 1991). Symptom development was observed daily. Plants showing hypersensitive reaction were discarded. Susceptible plants showing chlorosis, progressive necrosis were discarded as soon as detected. Only plants showing no symptoms or having a longer incubation period than Sel. 27 were maintained.

From 1979-1985 Dr. M. Zapata of the UPR in collaboration with Dr. R.Wilkinson of University of Cornell evaluated breeding lines under tropical conditions using inoculation with local strains of the pathogen at the UPR Fortuna Substation (Zapata et al., 1985). There was also some selection for resistance to ashy stem blight, as the fields had a high inoculum level of *Macrophomina phaseolina.* Seeds from plants selected for Xcp resistance were sent to Cornell for incorporation into the crossing program. The progenies of the crosses in the F_1 generation were returned to PR for evaluation.

In field plantings during five summer seasons (1986-1990) at the UPR Fortuna Substation, Dr. G. Freytag, USDA-ARS and Dr. M. Zapata inoculated with local Xcp strains and individual plant selections in the F_3 generation were made from heterogeneous lines selected for Xcp resistance. Plant rows from resistant plants were grown in nurseries during the winter season at the USDA-ARS Isabela Research farm and selected for plant habit and yield potential. Resistance on foliage of individual plants at flowering was confirmed three times by using multi-needle inoculations under controlled greenhouse environments at Mayagüez using 4 pure strains from the American Type Culture Collection (ATCC) and from two local sources. The results are presented in Table 2.

Table 1. Sources of resistance to *Xanthomonas campestris* pv. *phaseoli* used to develop the W-BB lines.

	Sources of resistance					
Lines	GN-1[1]	Sel. 27[2]	PI 207262[3]	PI 180745[4]	PI 180746[5]	65859[6]
W-BB-11	+[7]	+	+	+		
W-BB-20-1	+	+	+	+	+	+
W-BB-35	+	+	+	+	+	+
W-BB-52	+	+	+			+
W-BB-II-56	+	+	+		+	

[1]/GN-1 = Univ. of Idaho Great Northern #1.
[2]/Sel. 27 = Great Northern Nebraska #1 Selection 27.
[3]/207262 = Plant Introduction 207262 from Colombia, SA.
[4]/180745 = [5]/180746 = Plant Introduction (*P. coccineus* x *P. vulgaris*) from Germany.
[6]/65859 = (*P. vulgaris* x *P. coccineus*) from P.A. Lorz, Univ. of Florida.
[7]/+ = Indicates source of resistance present in the line.

Table 2. Reaction of individual bean lines to inoculation with *X. campestris pv. phaseoli* under greenhouse and field conditions.

	Greenhouse Xcp pathovar/origin					Field
Identitiy	*phaseoli* ATCC 9563	*phaseoli* PR 820	*fuscans* ATCC 11766	*vignicola* ATCC 11648	*glycines* ATCC 17915	*phaseoli* field strains PR
W-BB-11	I	I	T	I	I	T
W-BB-20-1	I	I	I	I	I	R
W-BB-35	I	I	S	I	I	T
W-BB-52	I	I	R	I	I	S
W-BB-II-56	I	I	I	I	I	R

I- Immune, no lesions; R = Resistance, very small (1-3mm), chlorotic but non-progressive lesions; T = Tolerant slow disease development, takes 8-10 days under controlled growth conditions and 44 days after inoculation under field conditions to develop 25% chlorotic lesions and less than 25% of necrotic lesions; S = Susceptible under controlled growth conditions takes 6 days to show symptoms and 44 days to develop necrosis on 50% of the inoculated tissue under field conditions.

Botanical Description

Line W-BB-11 (from Cornell line 84-4216-1) has a bushy vine (Type III) plant habit, a height of 70 cm, straight pods 5-7 cm long and small, gray to black seed weighing 0. 27g/ seed.

Line W-BB-20-1 (from Cornell line 84-4446-1) has a determinate bush (Type I) plant habit, a height of 40 cm, slightly curved pods 5-8 cm long and white seed weighing 0. 24g/seed. This line has the I gene resistance to Bean Common Mosaic Virus (BCMV).

Line W-BB-35 (from Cornell line 84-4454-1) has a determinate bush (Type I) plant habit, a height of 30 cm, late season, broad, straight pods 5-8 cm long and a large, rounded, yellow ship brown seed weighing 0.43g/seed. This line has a protected (probably by bc2-2) I gene resistance to BCMV.

Line W-BB-52 (from Cornell line 84-4610-3) has a determinate bush (Type I) plant habit, a height of 20 cm, early season, flat, curved pods 7-9 cm long with some snap characteristics and long, cream colored seeds weighing 0.30g/seed. This line does not have the I gene for BCMV resistance.

Line W-BB-11-56 (from Cornell line 85-8250-1) has a bushy vine (Type III) plant habit, a height of 70 to 120 cm, late season, curved pods 7-10 cm long with some snap characteristics and a good seed set in Puerto Rico of a pinto seed type weighing 0.25g/seed. This line has protected by bc2-^{2} I gene resistance to BCMV.

Identification of resistance using genetic markers

A portion of the resistance to common bacterial blight was derived from GN#1 Sel. 27 as indicated by the presence of the SCAR (sequence characterized amplified region) marker linked with a quantitative trait locus for resistance to common bacterial blight on linkage group B10 SAP-6-820 Miklas, 2000 et al., (Table 3). Suitable markers to identify the other sources of resistance have not been developed yet.

Table 3. Identification of bacterial blight resistance using four genetic markers (SCARs).

	SCARS Markers			
Lines	XAN 159 B-8 [1/] Su 91	OAC 88 B-8 R-7313	GN#1 Sel 27 B-10 SAP-6	XAN 159 B-6 BC 420
W-BB-11	-[2/]	-	+	-
W-BB-20-1	-	-	+	-
W-BB-35	-	-	+	-
W-BB-52	-	-	+	-
W-BB-11-56	-	-	+	-

[1/] Indicates the linkage group.

[2/] - Indicates absence and + presence of the marker for resistance

Seed availability

Seed of the germplasm lines from the F_8 generation is available upon request to the Bean Program, Tropical Agricultural Research Station, Agricultural Research Service, U.S. Department of Agriculture, P.O. Box 70, Mayagüez, Puerto Rico, 00681 or to Dr. Mildred Zapata, Crop Protection Department. University of Puerto Rico, P. O. Box 9030, Mayagüez, Puerto Rico, 00681-9030. We ask that appropriate recognition of source be given when this germplasm contributes to a new cultivar.

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