Five Multiple-virus-resistant Common Bean Breeding Lines

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The common bean (*Phaseolus vulgaris* L.) is threatened by >30 viral disorders worldwide. These diseases are primarily insect vectored, and host plant resistance provides the most effective and economical control method. The objective of our breeding program was to pyramid a series of resistance genes into a genetically diverse set of breeding lines. By developing an elite set of breeding lines with broad-spectrum virus resistance, a gene or group of genes can be more easily incorporated into existing regional cultivars. The specific viruses addressed by this effort include bean common mosaic (BCMV), bean yellow mosaic (BYMV), broad bean wilt (BBWV), blackeye cowpea mosaic (BICMV), clover yellow vein (CYVV), pea mosaic (PeMV),

soybean mosaic (SMV), tobacco mosaic (TMV), and watermelon mosaic virus (WMV).

Origin

These breeding lines were derived from a cross between Great Northern-1140 (GN-1140) and B-21 (Provvidenti et al., 1989). GN-1140 is resistant to BBWV (Provvidenti, 1983, 1988, 1993), CYVV (Provvidenti and Schroeder, 1973), PeMV (Schroeder and Provvidenti, 1968), SMV (Provvidenti et al., 1982), WMV (Provvidenti, 1974), and certain pathotypes of BCMV (Provvidenti, 1993) as conferred by *Bbw*, *cyv* (formerly *by-3*), *By*, *Smv*, *Wmv*, and *bc-u* coupled with *bc-I²*, respectively (Drijfhout, 1978; Kyle and Provvidenti, 1987;

Provvidenti, 1987a). B-21 carries resistance to BCMV, BYMV (Dickson and Natti, 1968), BICMV, cowpea aphid-borne mosaic virus (CABMV) (Provvidenti et al., 1983), TMV (Thompson et al., 1962), and WMV (Provvidenti, 1974) as governed by the *I*, *By-2*, *Bcm*, *Cam*, *Tm*, and *Hsw* genes, respectively (Kyle and Provvidenti, 1993; Provvidenti et al., 1989). B-21 also is resistant to PeMV (unpublished data). In B-21, the *I* gene confers resistance to BCMV pathogenicity groups I, II, IVa, Va, Vb, and VII and high-temperature-sensitive resistance to groups III, IVb, VIa, VIb (Drijfhout, 1978). The BCMV reaction profile of GN-1140 is equivalent to the differential GN-123, including tolerance to pathogenicity groups VIa and VIb; resistance to pathotypes I, II, III, Va, and Vb; and susceptibility to groups IVa, IVb, and VII as conditioned by *bc-u* and *bc-I²* (Table 1). When the *I* gene is coupled with these recessive alleles, tolerance or resistance is expanded to more BCMV pathotypes, and these genotypes are protected against systemic hypersensitive necrosis induced by high temperatures. The *I* gene also is linked tightly to a set of dominant genes that confers temperature-sensitive resistance to the potyviruses azuki mosaic virus (AZMV), BICMV, CABMV, and WMV and temperature-insensitive resistance to passionfruit woodiness virus-K (PWV-K) and zucchini yellow mosaic virus (ZYMV); however, infection with Thailand passionfruit virus (ThPV) and SMV produces a lethal veinal necrosis (Fisher and Kyle, 1995; Kyle and Dickson, 1987). Resistance to WMV in B-21 is governed by the temperature-sensitive *Hsw* gene and the seasonally affected *Wmv* gene from GN-1140 (Kyle and Provvidenti, 1987; Provvidenti et al., 1989). Resistance to SMV in GN-1140 is conferred by the temperature-insensitive *Smv* gene, but the *Hss* gene found in B-21 induces a temperature-insensitive lethal veinal necrosis (Table 1) (Kyle and Provvidenti, 1993). The *Wmv* and *Smv* genes cosegregate and are nonpathotype specific; *Hss* and *Hsw* also are nonpathotype specific and are linked tightly to *I*, as are *Bcm* and *Cam* (Table 1) (Fisher and Kyle, 1994; Kyle and Provvidenti, 1987, 1993). The previously listed

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Table 1. Virus diseases, resistance genes, sources of resistance, and isolates associated with the development of multiple-virus-resistant bean breeding lines.

Virus disease	Resistance gene(s)	Isolate	Source of resistance
Bean common mosaic (BCMV)	<i>I</i>	NY-15 (NY68-95)	B-21
	<i>bc-u</i> , <i>bc-I²</i>	NL-3	GN-1140 ^a
	<i>bc-u</i> , <i>bc-I²</i>	NL-5	GN-1140
	<i>bc-u</i> , <i>bc-I²</i>	NL-8 (NY82-20)	GN-1140
Bean yellow mosaic (BYMV)	<i>By-2</i>	Hagedorn <i>isol.</i> 1	B-21
Blackeye cowpea mosaic (BICMV)	<i>Bcm</i>	BICMV-Fla	B-21
Broad bean wilt (BBWV)	<i>Bbw</i>	NY82-30	GN-1140
Cowpea aphid borne mosaic (CABMV)	<i>Cam</i>	CABMV-Mor	B-21
Clover yellow vein (CYVV)	<i>cyv</i>	NY68-104	GN-1140
Pea mosaic (PeMV)	<i>By</i>	NY58-1027	GN-1140
Soybean mosaic (SMV)	<i>Smv</i>	NY76-6	GN-1140
Tobacco mosaic (TMV)	<i>Tm</i>	McR & Alex-3	B-21
Watermelon mosaic (WMV) ^b	<i>Wmv</i>	NY62-76	GN-1140
	<i>Hsw</i>		B-21

^aGN-1140 = Great Northern 1140.

^bFormerly watermelon mosaic virus-2 (WMV-2).

diseases are potyviruses, except for BBWV, a fabavirus, and TMV, a tobamovirus (Provvidenti, 1993).

A modified single-seed descent (SSD) program was used to develop five breeding lines (Brim, 1966; Scully and Federer, 1993). For simply inherited traits, SSD schemes can effectively stack desirable genes from the donor parents to produce recombinant inbred lines. This SSD program was modified to include selection for resistance to a single different virus with each generation of inbreeding, the use of double-seed descent in the F_3 generation, and a delay of progeny testing until the conclusion of all selections (Fig. 1). In the F_2 generation, 1400 plants were inoculated with CYVV to reveal the homozygous *cyv* individuals. Two progeny from each of the CYVV-resistant plants were advanced to the F_3 and inoculated with the African strain NL-8 (pathogenicity group III) of BCMV (Provvidenti et al., 1984) to reveal the double recessive *bc-u/bc-u bc-l²/bc-l²* individuals. Recessive genes were screened in the F_2 and F_3 generations in an effort to reduce the frequency of heterozygous loci in the F_4 to F_7 , where dominant genes were selected (Table 1, Fig. 1). F_3 -derived single progenies were carried forward to the F_4 and challenged with BYMV. Single F_5 progenies then were taken from BYMV-resistant F_4 plants and inoculated with the NY-15 strain (pathogenicity group Va) of BCMV. This process was repeated in the F_6 and F_7 generations for BBWV and PeMV, respectively (Fig. 1). In the six generations (F_2 to F_7) of selection, seedlings were inoculated mechanically at least twice using standard practices and isolates of each pathogen (Table 1). Seventeen F_2 -derived F_7 breeding lines survived the selection phase and advanced for progeny testing to the six viruses or pathotypes. In addition, these breeding lines were submitted to a second set of inoculations with BICMV, SMV, TMV (McRitchie and Alexander, 1963), WMV, and the necrosis-inducing strains NL-3 and NL-5 (pathotypes VIa and VIb, respectively) of BCMV (Drijfhout, 1978). Based on the genetics of the parents, resistance to some of these diseases was expected in the progeny. The *I* gene was screened in F_5 ; thus, resistance to diseases associated with *I* (Lamprecht's linkage group III) also was expected (Table 2) (Kyle and Dickson, 1987). After progeny testing, five of the 17 lines were chosen for release, each with a different seed color.

Description

Virus reactions. Resistance is defined by three criteria: 1) the absence of symptoms and the virus is not recoverable from new tip growth; 2) the presence of small local chlorotic or necrotic lesions around the infection site; and 3) "tolerance," where plants initially show mild or transient symptoms of systemic mottle, but the new growth has cleared symptoms. The two observed susceptible reactions are apical necrosis, or systemic mosaic and epinasty. For the pathotypes of BCMV, all five breeding lines exhibited resistance to NL-3, NL-8, and NY-15 but varied for NL-5

(Table 2). B-21, the source of the *I* gene, developed apical necrosis due to breakdown of resistance, which was attributed to elevated temperatures ($\geq 30^\circ\text{C}$) in the greenhouse. These lines also were resistant to BBWV, BICMV, BYMV, CYVV, PeMV, SMV, TMV, and WMV. WMV inoculations were not conducted across a temperature differential, so resistance based on *Hsw* or *Hsw* in combination with *Wmv* could not be distinguished specifically.

Morphology. These five breeding lines produce indeterminate plants that range to 50 cm in canopy height and spread, and generally carry 80 to >100 leaves on a CIAT Type IIb plant growth habit (Adams et al., 1985). Plants are somewhat open, irregularly rounded, and more or less upright, but they become semiprostrate as the pod load matures. The canopy is distinguished by a more compact appearance toward the base, with four to seven protruding runners that commonly range from 50 to 75 cm but may extend ≤ 1.0 m and intertwine variably. The main stem has ≤ 20 nodes, with secondary branching on nodes five through eight. Typically, the first and last secondary branches are short, with only four to six nodes, but the predominant secondary branches have 10 to 15 nodes and ≤ 20 cm internode distances. Tertiary branches are absent. This morphology is based on plants grown in upstate New York (42°N) and the High Plains of Texas (38°N).

The first flowers are set between nodes five and eight, with anthesis occurring after the expression of nine to 11 main-stem nodes. Flowers are produced up the main stem and on secondary branches, but fruit set ceases before main-stem node 15. Most pods are set within the canopy, and all are indehiscent types typical of dry beans; however, some lines have invaginated locules.

SP 6C. Seeds of this "off-type" pinto have a light tan to brown background with black foreground markings, and a usual test weight of 25.0 g. Individual seed average 1.13 cm long, 0.65 cm wide, and 0.50 cm deep; they are slightly curved to straight. Pods range to 13.0 cm in length and 1.0 cm in width. They are straight to slightly bowed, partially undulate, and round to flat. The apicule is curved and 1.0 to 1.5 cm long. Pods have five to seven seeds, dry to a tan color, and are prominently streaked purple, which eventually fades to brown. A single inflorescence with four to six purple flowers is produced in each fertile axil having about two pods set per inflorescence. SP 6C has suffused purple throughout the stems as well as the cotyledons, hypocotyl, sepals, and veins.

SP 17B. Seeds of this genotype are white with visible veins, transversely round to oblong, and laterally straight. Seed dimensions average 1.15 cm long, 0.71 cm wide, and 0.53 cm deep, with a test weight of 23.0 g. Pods are

Fig. 1. A modified single-seed descent scheme used in the development of five virus-resistant common bean (*Phaseolus vulgaris* L.) lines.

Table 2. Reaction profile of five breeding lines and standard cultivars to an array of viral diseases.²

Germplasm	Virus ³											
	BCMV				BYMV	CYVV	BBWV	PeMV	BICMV	SMV	TMV	WMV
	NY-15	NL-8	NL-3	NL-5								
<i>Breeding lines</i>												
SP 6C	R	R	R	SN/R	R	R	R	R	R	R	R	R
SP 17B	R	R	R	R	R	R	R	R	R	R	R	R
SP 61	R	R	R	SN	R	R	RL	R	R	R	RL	R
SP 180D	R	R	R	R	R	R	RL	R	RL	RL	RL	R
SP 377C	R	R	R/RL	SN/R	R	R	RL	R	R	R	RL	R
<i>Standards</i>												
B-21	SN*	SN	SN	SN	R	SN	SN	R	R	SN	RL	R
GN-1140	R*	R	R*	R*	SM	R	RL	R	RL	R	RL	R
GN-31	---	---	R	R	---	---	---	---	---	---	---	---
Red Kidney	SM	R	SM	SM	SM	SM	---	---	---	---	---	---
Black Turtle-1	R	SN	SN	SN	SM	SM	---	---	---	---	---	---
Black Turtle-2	SM	SM	SM	SM	SM	SM	---	---	---	---	---	---
Bonneville Pea	---	---	---	---	R	R	---	---	---	---	---	---
Ranger Pea	---	---	---	---	SM	SM	---	---	---	---	---	---

R = resistant with no symptoms; RL = resistant with small local mosaic or necrotic lesions; R = tolerance, plants initially developed systemic mottle, but later clear symptoms; SN = susceptible with apical necrosis; SM = susceptible with systemic mosaic.

²BCMV = bean common mosaic, BYMV = bean yellow mosaic, CYVV = clover yellow vein, BBWV = broad bean wilt, PeMV = pea mosaic, BICMV = blackeye cowpea mosaic, SMV = soybean mosaic, TMV = tobacco mosaic, and WMV = watermelon mosaic virus.

³B-21 is the source of the *I* gene, which confers resistance to NY-15. Apical necrosis is due to the high-temperature sensitivity of this gene.

mostly 9.0 to 12.0 cm long and 0.8 to 1.2 cm wide. Their shape is flat, somewhat undulate, partially constricted between locules, and usually arched ≤ 2.0 cm. The apicule is curved and 1.0 to 1.2 cm long, with a slight hook. Five to seven seeds are loosely encased in tan-colored dry pods with faint surface markings. Two to four pods develop on a single axillary inflorescence with four to six white flowers. A second rudimentary inflorescence rarely develops. Peduncles and pedicels range from 1.0 to 6.0 cm and 0.3 to 0.6 cm long, respectively.

SP 61B. This genotype has brown to tan seeds with visible veins on the testa. Seeds are longitudinally straight to slightly curved and somewhat flattened transversely. Commonly, seeds have a test weight of 28.4 g, with a seed length 1.25 cm, a width of 0.71 cm, and a depth of 0.49 cm. Pods are flat, undulate, partially constricted, and arched ≤ 2.5 cm, and they range from 10.5 to 12.5 cm in length and to ≈ 1.0 cm in width. Pods dry to a tan color, but they may have a faint mauve or cranberry tint. Mostly, five seeds are held loosely in pods that have four to six locules. Of the two to four flowers produced in a dominant axillary inflorescence, one to three set pods. A second axillary inflorescence rarely sets fruit. Flowers are white, but the banner is suffused with light purple at the base. SP 61B was the earliest of these lines, often flowering at the fourth or fifth main-stem node.

SP 180D. This genotype has a test weight of 23 g, and the seed averages 1.11 cm long, 0.70 cm wide, and 0.46 cm deep. Seeds are dull black, straight to slightly curved, and transversely rounded to partially flattened, with a dimpled testa. Pods mostly range from 10 to 12 cm in length and 0.9 to 1.1 cm in width, with a curved apex that extends ≤ 1.3 cm and is sometimes hooked. Pods range from nearly straight to a 2.0-cm arch that is shifted toward the apex. Pods are flattened, partially undulate, and constricted. Six seeds are usually set in pods that have five to seven locules. Over time, pods dry to a tan color and are tinted

purple. Three to four purple flowers are produced in a single inflorescence, which elongates to ≤ 12 cm within the canopy, but commonly average 5 cm and sets pods on about half the flowers. A second rudimentary inflorescence is occasionally expressed but often aborts.

SP 377C. This pinto breeding line produces seeds that average 1.14 cm long, 0.70 cm wide, and 0.51 cm deep, with a test weight of 27 g. Seeds are mostly straight and transversely flattened to round. Pod length ranges from 8 to 11 cm and varies from 0.70 to just >1.0 cm in width. Pod shape is usually flat but sometimes rounded, occasionally undulate and arched ≤ 1.0 cm, with a curved and hooked apex that ranges between 0.50 and 1.0 cm long. Five to six seeds are loosely enclosed in tan pods that are faintly streaked. Two inflorescences are set in fertile axils. The dominant inflorescence produces two to six flowers, but the secondary inflorescence sets two flowers at most. About half of the flowers set pods. Flowers are white with mauve streaks in the banner.

Availability

Small quantities of greenhouse-produced seed are available from B.S. Field-produced seed also is available. These breeding lines are released without restriction.

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