

## Bean Common Mosaic Virus - BCMV & Bean Common Mosaic Necrosis Virus - BCMNV

Bean common mosaic virus (BCMV) and bean common mosaic necrotic virus (BCMNV) are aphid-transmitted potyviruses that can cause significant bean yield loss (Gálvez and Morales, 1989). This seed-borne virus can persist in infected seed lots.

There are two major forms of resistance: race-specific genes that require *bc-u* for expression, and the dominant *I* gene. Five alleles at three loci condition resistance to BCMV and BCMNV. Drijfhout's (1978) tables 6 and 31 provide information on isolate – host compatibility reactions. With the exception of *bc-3*, race specific genes provide resistance to some but not all pathogenicity groups of the viruses. Susceptible cultivars show vein banding and mosaic mottle symptoms in their leaves, and may allow seed transmission. Plant with resistance to a specific isolate generally remain symptomless, although some host – pathogen combinations will produce necrotic local lesions. The *I* gene gives immunity to BCMV isolates at temperatures less than 30C. At temperatures greater than 30C and for BCMNV isolates infecting at any growing temperature, a plant will first exhibit pinpoint necrotic local lesions, followed by veinal necrosis and eventually top necrosis, which kills the plant. This symptom is called “black root” because of the blackened necrotic veins apparent when stems, pods and roots are cross-sectioned. When the race-specific genes are combined with the *I* gene, they will “protect” the *I* gene from necrosis inducing isolates for which they provide specific resistance. The viruses are never seed-transmitted in cultivars possessing the *I* gene, but fields of an unprotected *I* gene cultivar may suffer total crops failure if a susceptible cultivar harboring BCMNV isolates is planted in close enough proximity for aphids to transport the virus from the susceptible to the *I* gene host.



Black root

Mosaic

Photograph provided by H. F. Schwartz  
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Mills and Silbernagel (1992) described a protocol for preparation of inoculum and inoculation of bean plants with BCMNV. The BCMNV can be maintained on the susceptible cultivar 'Sutter Pink' or 'Dubble Witte' (The latter is a better host for certain isolates). Approximately 25% of the seed harvested from BCMNV infected Sutter Pink plants are often infected with the virus (J. Kelly, personal communication). However, percent seed transmission is affected by time of inoculation; earlier inoculation causes higher seed transmission from emergence to bloom. Seed transmission will be very low or nonexistent after flowering). Infected seed can be used to store the virus when not conducting inoculations. Inoculum can also be stored in infected leaf tissue that has been lyophilized, or in fresh collected tissue stored in a -80C freezer.

Inoculum is prepared by grinding 1 g young BCMNV infected leaves in 5 ml of cold 0.01 M phosphate buffer, pH 7. Primary leaves are inoculated when about  $\frac{3}{4}$  expanded (7-8) days after planting. Leaves were dusted with 600 mesh carborundum and the inoculum was gently rubbed on the entire surface of the primary leaf. Carborundum can also be incorporated into the virus-buffer mixture. With unprotected *I* gene and BCMNV isolates, necrotic local lesions may appear in as few as 3 days with top necrosis ensuing within a week (the reaction is temperature dependent, and may take up to a week for symptoms to appear in a cool environment). Certain isolate – host cultivar combinations such as NL-3 isolate inoculated into a plant with *I bc-1<sup>2</sup>* will show delayed, symptoms. Symptoms in the absence of the *I* gene will take two to three weeks to appear (symptoms are typically read at 21 – 28 days).

Large scale field inoculations can be done with an airless electric paint sprayer. Inoculum is made up in liter batches with the buffer solution as described above. It is critical to use chilled buffer for tissue maceration, and the inoculum should be kept on ice until use. Approximately 100 g of infected leaf tissue is placed in an industrial blender with a liter of buffer. Tissue is macerated on the high setting for three minutes, and then strained through three layers of cheesecloth. The inoculum is applied through the paint sprayer without carborundum. Depending on the type of sprayer, it may be necessary to remove the nozzle guard to allow the nozzle of the sprayer close enough to the plants to achieve a water-soaked lesion when the sprayer is activated. This method allows the inoculation of a hundred meters of row in about a half hour with about 95% of susceptible inoculated plants showing symptoms in four weeks. Aphids will generally continue the spread of the virus in the field such that all susceptible plants will eventually show symptoms.

Mills and Silbernagel (1992) proposed a 1-9 evaluation scale (Table 1) in accordance with the scale proposed by the CIAT Bean Research Program where 1-3 is considered resistant, 4-6 intermediate and 7-9 are susceptible. An alternative 0-10 evaluation scale for BCMNV reactions was proposed by Strausbaugh et al. (2003a).

The NL3 strain on BCMNV can be used to screen bean lines for resistance to both BCMV and BCMNV. Kelly (1997) described the differential host reaction of eight varieties of common bean inoculated with the NL3 strain of BCMNV (Table 2). Cultivars such as 'Raven' that combine the *I* and *bc-3* genes are resistant to all known strains of BCMNV and BCMV (Kelly et al., 1994). Most commercial seed types of common bean have at least one line with the *I* and *bc-3* resistance genes (Table 3).

Care must be taken when evaluating the NL3 – *bc-1*<sup>2</sup> host group reactions because some symptomless plants will have high virus titer in their tissues (detectable by ELISA) and are capable of seed transmission (Strausbaugh et al., 2003b).

Enzyme-linked immunosorbent assay (ELISA) provides a means of detecting infection by BCMV and BCMNV. A broad-spectrum polyclonal antibody that detects most potyviruses, a monoclonal that is specific to BCMV and BCMNV, and a monoclonal that is specific to BCMNV have been developed. ELISA kits are available commercially from Agdia (<http://www.agdia.com/>).

Table 1. Rating scale (1-9) used to evaluate beans for reaction to bean common mosaic necrotic virus (BCMNV) strain NL3.

BCMNV score	Primary leaf inoculation <sup>1</sup>	
	Inoculated leaf reaction	Systemic reaction
1	None	None
2	Few to many restricted necrotic local lesions (NLL)	None
3	Few to many NLL with slightly spreading veinal necrosis (SVN)	None
4	Few to many NLL with SVN	Vascular necrosis (VN) spread from main vein to primary leaf petiole.
5	Few to many NLL with SVN	VN spreads beyond primary leaf petiole into main stem of plant.
6	Few to many NLL with SVN	Vein spreads from stems and branches
7	Few to many NLL with SVN	VN on trifoliolate leaves coalesces, leaves drying.
8	Few to many NLL with SVN	VN spreads killing one or more branches of the plant
9	Few to many NLL with SVN	VN kills entire plant

<sup>1</sup> Inoculum prepared at a rate of 1 g infected leaf tissue titrated on 5 ml cold 0.01 M phosphate buffer, pH 7. Inoculum was rubbed on a  $\frac{3}{4}$  expended primary leaves, about 8 days after planting at 27° C.

Source: Mills and Silbernagel (1992).

Table 2. The differential host reaction of eight varieties of common bean inoculated with the NL3 strain of BCMNV.

Variety	Genotype <sup>1</sup>	Host symptoms
Sutter Pink	<i>ii</i>	M – Mosaic
Black Turtle Soup 1	<i>ll</i>	TN – Top necrosis
Olathe	<i>ii bc-1<sup>2</sup>bc-1<sup>2</sup></i>	MM – Mild mosaic
Beryl	<i>ll bc-1<sup>2</sup>bc-1<sup>2</sup></i>	VN – Vein necrosis
Othello	<i>ii bc-2<sup>2</sup>bc-2<sup>2</sup></i>	NR – No reaction
92US-1006	<i>ll bc-2<sup>2</sup>bc-2<sup>2</sup></i>	NLL – Necrotic local lesions
G94574	<i>ii bc-3bc-3</i>	NR – No reaction
Raven	<i>ll bc-3bc-3</i>	NR – No reaction

<sup>1</sup> The presence of the *bc-u* is assumed in those varieties with specific *bc* genes but is not shown.

Source: Kelly (1997).

Table 3. Sources of resistance to BCMV and BCMNV in different seed classes.

Name or number	Seed color / type	Resistance genes	Reference
Raven	9 / Black	// <i>bc-3bc-3</i>	Kelly et. al (1994)
BelMiDak RMR 10-12	1 / Navy	// <i>bc-3bc-3</i>	Pastor-Corrales (2003)
BelDakMi RMR 19-23	2M / Pinto	//, <i>bc-3bc-3</i>	Pastor-Corrales (2003)
BelMiNeb RMR 9-13	1 / Great Northern	// <i>bc-3bc-3</i>	Pastor-Corrales (2003)
Merlot	7 / Red Mexican	<i>ii bc-1<sup>2</sup> bc-1<sup>2</sup></i>	Hosfield et al. (2004)
PR9357-107	6 / Small red	// <i>bc-3bc-3</i>	Beaver et al. (1998)
USWA-61 & 63	5 / Pink	// <i>bc-1bc-1</i> or // <i>bc1<sup>2</sup>bc1<sup>2</sup></i>	Miklas (1997a)
USCR 7	2R / Cranberry	// <i>bc-3bc-3</i>	Miklas (1998a)
BRB 198	6M / Red mottled	// <i>bc-3bc-3</i>	
Rojo USDK-4 to 6	6K / Dark red kidney “	// <i>bc-3bc-3</i> “	<hr/> Miklas (1998b)
USLK-1-3	5K / Light red kidney	“	Miklas (1998c)
USWA 64 & 68	1 / Snap	// <i>bc-1bc-1</i>	Miklas (1997b)

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