Bean Golden Yellow Mosaic Virus

Bean golden yellow mosaic (BGYMV), a whitefly (*Bemisia tabaci*) transmitted gemnivirus, is a serious bean disease in Central America, the Caribbean and southern Florida (Blair et al., 1995; Gálvez and Morales, 1989). Significant progress has been made toward the development of cultivars and breeding lines with resistance to BGYMV. (Beaver et al., 2003; Beebe, 1994).

Replicated field trials were successfully used in Guatemala to screen bean lines for resistance to BGYMV (Morales, 1994). Spreader rows should be planted at least three weeks before the nursery is planted. A susceptible variety of common bean or lima bean (*Phaseolus lunatus* L.) can be used to increase BGYMV whereas soybeans ([*Glycine max* L. (Merr.)] can be used to increase whitefly populations. Frequent applications of pesticide on the spreader rows were used in Puerto Rico to suppress the populations of the natural enemies of the whiteflies. A BGYMV susceptible bean line should be planted every few rows to monitor disease pressure. In Puerto Rico, leaf chlorosis was evaluated at 30 and 45 days after planting whereas pod deformation was evaluated at 45 and 60 days after planting (Acevedo Román et al., 2004; Osorno et al., 2003).

Artificial inoculation has also been used at the Centro Internacional de Agricultura Tropical (CIAT) in Colombia to screen bean lines in the greenhouse for BGYMV resistance (Morales,1987). Dry infected tissue or fresh tissue can be used to prepare the inoculum. Bean plants should be inoculated at the primary leaf stage, approximately 7-10 days after planting. Dry infected tissue should be hydrated for 10 minutes in a buffer solution (potassium phosphate 0.1 M, pH 7.5) before being finely ground in a mortar (Table 1). These steps should be conducted under cold temperatures by surrounding the equipment with ice. The proportion of fresh tissue to buffer tissue (w:v) should be close to 1:4. The process of hydrating the tissue is facilitated by determining the weight before drying. A small amount of a fine abrasive can be applied to the primary leaves

before inoculation. Cotton swabs can be used to inoculate the leaf by gently rubbing the swab with inoculum on the surface of the leaf. *The inoculum should be used within 15 minutes after preparation*. Inoculated plants should be kept under moderate light and temperatures during the first 24 h after inoculation. After the first day, plants can be moved to full sunlight and a temperature near 30° C.

Table 1. Preparation of 0.1 M phosphate buffer.

0.2 M Monobasic Stock	Combine 13.9 g sodium phosphate monobasic in 500 mL distilled H_20	
0.2 M Dibasic Stock	Combine 53.65 g sodium phosphate dibasic heptahydrate (or 28.4 g of the anhydrous form) in 1 L distilled H_20	
0.1 M Buffer	Combine 600 ml distilled H_2O and the appropriate amounts of monobasic and dibasic stock to obtain the desired pH, according to the following table:	
Monobasic stock	*Dibasic stock*	*pH*
(ml)	*(ml)*	
205.5	*94.5*	*6.5*
187.5	*112.5*	*6.6*
169.5	*130.5*	*6.7*
153	*147*	*6.8*
135	*165*	*6.9*
117	*183*	*7.0*
99	*201*	*7.1*
84	*216*	*7.2*
69	*231*	*7.3*
57	*243*	*7.4*
48	*252*	*7.5*
39	*261*	*7.6*
31.5	*271.5*	*7.7*
25.5	*274.5*	*7.8*
21	*279*	*7.9*
15.9	*284.1*	*8.0*

Source: www.msu.edu/user/eisthen/lab/methods/anatomy/recipes/PB.html

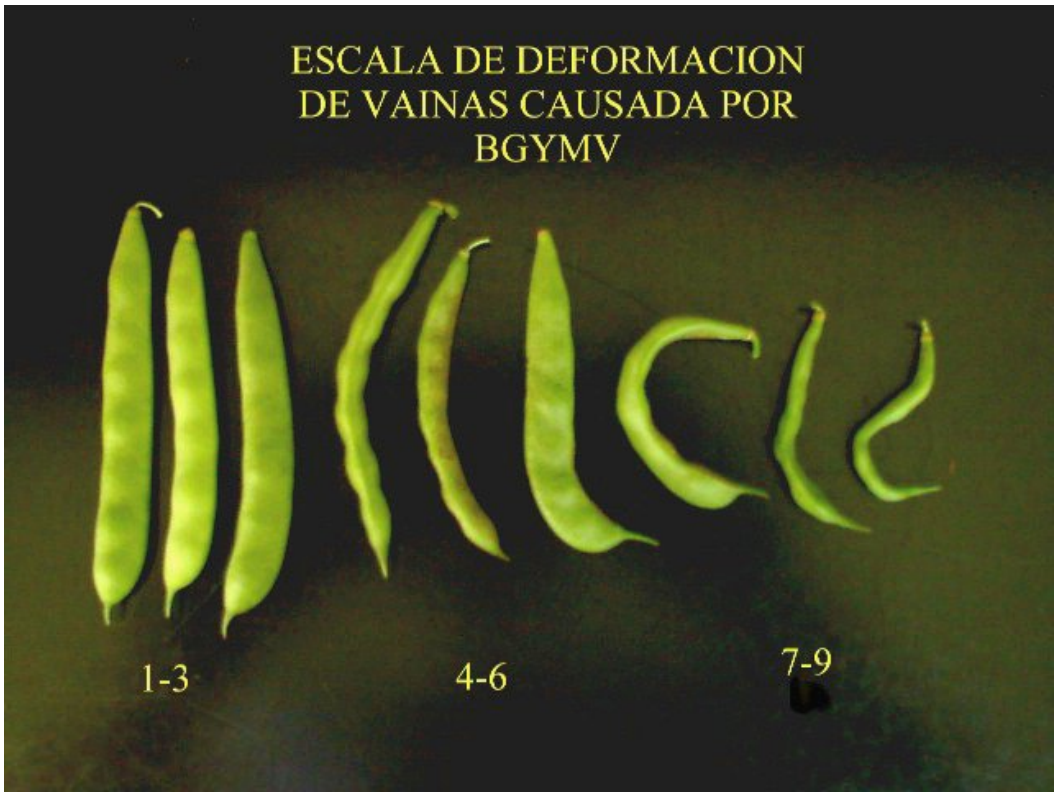
Morales (1987) also developed techniques for the isolation and conservation of BGYMV. The virus was reported to be active up to three months by keeping the dried tissue in a refrigerator. The virus should be isolated from an expanding leaf from a young plant with BGYMV symptoms. The infected leaf should be sealed into a small container containing a dessicant. Only a small amount of leaf tissue should be incorporated into the container in order to not exceed the capacity of the container to absorb moisture. If humidity appears within the

container after 24 h, the leaf tissue should be transferred to a new container with unused desiccant.
The dried leaf tissue should be maintained in a refrigerator.

Adames-Mora et al. (1996) developed a technique using whiteflies for screening beans in the greenhouse for resistance to BGYMV. Aviruliferous whiteflies were reared on soybeans Soybeans were planted every 21 days to maintain the whitefly colony. Irrigation on the oldest soybean plants were suspended to encourage the whiteflies to feed and lay eggs on younger soybean plants. The susceptible cultivar 'Top Crop' with typical BGYM symptoms, approximately 20 days after their inoculation, was used as the source of BGYMV. Aviruliferous whiteflies were placed in a cage for 72 h with BGMV infected bean plants (14 days after inoculation) to acquire the virus. The cage was covered with a black cloth during the first six hours of acquisition to avoid attraction of whiteflies to the light. After 72 h, twelve viruliferous whiteflies were placed in a small cylindrical cage covering a bean plant in a pot. The cage, which was made from a PVC tube, was 11 cm in diameter and 18 cm in length. Polyester covers were glued to the windows of the cages (Durazo and Natwick,1985). The whiteflies were collected early in morning when the insects are more sedentary. To collect the whiteflies, a plastic dropping pipet was attached to a 50 cm long plantic tube with an interior diameter of 0.2 cm. A 62 mesh (0.025 cm^2) was placed between the dropping pipet and the plastic tube to prevent the whiteflies from entering the tube. Suction was applied to the other end of the tube to collect the appropriate number of whiteflies. The whiteflies entered the cages through an 0.5 mm diameter orifice that was taped shut after the flied were introduced. After feeding on the bean plants for 72 h, the whiteflies were killed with an insecticide.

Inoculating plants with viruliferous white flies

Symptoms can appear on susceptible plants as soon as 1 week after inoculation. Adames-Mora et. al. (1996) found that some lines, such as DOR 364, with quantitative resistance, have delayed (up to 5 days) symptom expression. Lines homozygous for resistance genes *bgm-1* or *bgm-2* do not express chlorosis when inoculated with BGYMV (Velez et al. 1998) whereas lines with the *Bgp-1* gene (Acevedo-Román et al., 2004) do not develop deformed pods in the presence of BGYMV. Osorno et al. (2003) reported that two genes from *P. coccineus* confers resistance to BGYMV: a recessive gene provides resistance to chlorosis and a dominant gene confers resistance to pod deformation. The differential host reaction of common bean lines inoculated with BGYMV are described in Table 2. Most of the seed classes of beans produced in the tropics have at least one BGYMV resistant.line or cultivar (Table 3).



Scale (1-9) used in Puerto Rico to evaluate pod deformation caused by BGYMV (Acevedo Román et al, 2004).

Table 2. Different host reactions of common bean lines inoculated with BGYMV.

Line	Genotype	Host symptoms
Top Crop	*Bgm-1Bgm-1*	Mosaic and deformed pods
DOR 364	QTLs for resistance	Delayed symptom expression
A 429	*bgm-1bgm-1*	No mosaic, deformed pods
DOR 303	*bgm-2bgm-2*	No mosaic, deformed pods
Morales, Tio Canela 75	*bgm-1bgm-1 Bgp-1Bgp-1*	No mosaic, no deformed pods

Table 3. Sources of resistance to BGYMV in different seed classes.

Name or number	Seed color / type	Resistance genes	Reference
Turbo III, ICTA Ligera PR0247-49	9 / Black	*bgm-1* P. coccineus	Beaver et al. (2004)
Morales	1 / White	*bgm-1, Bgp-1*	Beaver et al. (1999a)
	2M / Pinto	*bgm-1*	
	1 / Great Northern		
	7 / Red Mexican		
Tio Canela 75 PR9771-3-2	6 / Small red	*bgm-1, Bgp-1* P. coccineus	Rosas et al. (1997) Beaver et al. (2005)
Rosada Nativa	5 / Pink	*bgm-1, Bgp-1*	Beaver et. al (1999b)
	2R / Cranberry		
PR9745-232, RMC-3	6M / Red mottled	*bgm-1*	Blair et al. (2004)
PR9443-4	5K / Light red kidney	*bgm-2*	Beaver et al. (1999c)
BelDade RGMR 4,5,and 6 (Indeterminate)	1 / Snap	*bgm-1*	Stavely et al. (2001)

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