Susceptibility of *Rudbeckia, Zinnia, Ageratum,* and Other Bedding Plants to Bidens Mottle Virus

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ABSTRACT

Logan, A. E., Zettler, F. W., and Christie, S. R. 1984. Susceptibility of *Rudbeckia, Zinnia, Ageratum,* and other bedding plants to bidens mottle virus. Plant Disease 68: 260-262.

Bidens mottle virus (BMoV) was identified as a pathogen of naturally infected plants of *Rudbeckia hirta hybrida, Zinnia elegans,* and *Ageratum conyzoides* (family Compositae) growing as bedding plants in Florida. Eight other composite bedding plant species and *Petunia hybrida* (Solanaceae) and *Verbena hybrida* (Verbenaceae) were also susceptible to this virus when manually inoculated.

Additional key words: *Calendula officinalis, Callistephus chinensis, Centantherum intermedium, Dimorphotheca pluvialis, Gaillardia grandiflora, Helianthus annuus, Helichrysum bracteatum, Stokesia laevis*

Bedding plants are becoming increasingly popular nationwide, and in Florida alone, they constitute an $8 million industry (1). Despite their importance, the virus diseases of bedding plants have received little or no attention. This paper reports natural infection of *Rudbeckia hirta hybridum, Zinnia elegans,* and *Ageratum conyzoides* by bidens mottle virus (BMoV) in Florida and assesses the susceptibility of other common bedding plants to this virus.

The electron microscope used in this research was purchased in part by funds from National Science Foundation Grant PCM-7825524.

Florida Agricultural Experiment Stations Journal Series No. 4971.

Accepted for publication 21 November 1983.

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MATERIALS AND METHODS

Plants were inoculated manually with leaves triturated in either distilled water or 0.01 M phosphate buffer (pH 7.6) and applied to test plants previously dusted with 600-mesh Carborundum. With the exception of *Stokesia laevis*, all inoculated plants were grown from seed and maintained in greenhouses at 24–28 C. The isolate of bidens mottle virus (BMoV-R) used throughout this study was isolated in spring of 1982 from perenniating plants of *Rudbeckia* growing in Orange County, FL. An isolate of BMoV from endive (BMoV-E) (10) was used for comparison in the transmission trials. Both isolates were maintained in *Nicotiana* × *edwardsonii* (4). Seedlings of *Chenopodium amaranticolor* and/or *Z. elegans* (9) were included in the inoculation experiments to ascertain inoculum viability and in attempts to recover the virus from inoculated plants, regardless of whether symptoms were expressed.

Epidermal strips taken from the lower leaf surface of infected *N.* × *edwardsonii* and *R. hirta hybridum* plants were stained in calcomine orange and Luxol brilliant green (2) and examined with the light microscope for inclusion bodies.

Leaf extracts for electron microscopy were prepared by dicing tissue in 0.01 M potassium phosphate buffer (pH 6.8). The extracts were transferred to carbon-coated grids and allowed to remain for 1 min. The grids were washed sequentially with 0.01 M potassium phosphate buffer, distilled water, and a 2% aqueous solution of uranyl acetate containing 125 μg/ml of bacitracin (7). The grids were then examined for viral particles with a Hitachi H-600 electron microscope.

Double radial immunodiffusion tests were done in petri plates containing 0.8% Noble agar, 1% NaN_3, and 0.5% sodium dodecyl sulfate (8). BMoV-E (10), the endive isolate, and BMoV-F, an isolate from *Fittonia* (11), were serologically compared with BMoV-R. Antisera to BMoV-E (provided by D. E. Purcifull, Department of Plant Pathology, University of Florida, Gainesville) and BMoV-F were used.

RESULTS

Twelve of the 31 species of bedding plants were susceptible to BMoV-R when inoculated manually (Table 1). In addition, this virus also infected *Bidens bipinnata, N.* × *edwardsonii,* and *C. amaranticolor.* Symptoms varied considerably among the inoculated plants and ranged from severe mottling, leaf distortion, and vein necrosis in *Calendula officinalis* to no apparent foliar symptoms in *Gaillardia grandiflora* (Table 1). Symptoms of *R. hirta hybridum*

seedlings inoculated with either BMoV-R or BMoV-E closely resembled those noted on the plants from which BMoV-R was originally obtained (Fig. 1). Systemic infections were obtained in all susceptible plants except *C. amaranticolor*, which reacted with local lesions as described previously for BMoV (3). The plants that did not become infected were *Dahlia variabilis* 'Rainbow,' *Chrysanthemum maximum* 'Alaska,' *Chrysanthemum carinatum, Cosmos bipinnatus* 'Sensation,' *Gerbera jamesonii, Tagetes patula* 'Petite,' *Antirrhinum majus* 'Cherrio,' *Impatiens balsamina* 'Double Camellia Flowered,' *Impatiens walleriana* 'Semi-dwarf,' *Viola tricolor* 'Swiss,' *Vinca rosea, Tropaeolum majus* 'Cherry Rose,' *Amaranthus hybridus* 'Early Splendor,' *Celosia argentea, Dianthus caryophyllus* 'Dwarf Fragrance,' *Portulaca grandiflora, Salvia splendens* 'Bonfire,' *Coleus blumei* 'Rainbow,' *Lobularia maritina* 'Carpet of Snow,' *Matthiola incana*, and *Limonium sinuatum*.

Table 1. Susceptibility of bedding plants to an isolate of bidens mottle virus from *Rudbeckia*

Plant	Symptoms[a]	Test[b]
Compositae		
Ageratum conyzoides 'Blue Mink'[c]	S,D,A	1,2,3
Calendula officinalis 'Orange Cornet'[c]	S,M,D,V	1,2,3
Callistephus chinensis 'Crego'[c]	D	2,3
Dimorphotheca pluvialis[c]	S,D	1,2,3
Gaillardia grandiflora[c]	L	2,3
Helichrysum bracteatum[c]	S,D,M,T	1,2,3
Helianthus annuus 'Teddy Bear'	S,D,M,A	2,3
Stokesia laevis[c,d]	M	2,3
Rudbeckia hirta hybrida[c,d]	S,M,D,C,A	1,2,3
Zinnia elegans	S,M,D,C	1,2,3
Solanaceae		
Petunia hybrida 'Confetti'	S,M,D,C	1,2,3
Verbenaceae		
Verbena hybrida 'Starlight'[c]	S,M,D	1,2,3

[a] S = stunting, M = mottling, D = leaf distortion, V = vein necrosis, T = leaf tip necrosis, L = latent, C = color break in flowers, and A = flower abortion and/or deformation.
[b] 1 = infections confirmed by detecting flexuous-rod shaped particles in negatively stained leaf extracts, 2 = immunodiffusion tests against BMoV-E and BMoV-F antisera, and 3 = inoculations to *Zinnia elegans*.
[c] Plants not previously tested for susceptibility to BMoV.
[d] Perennial species.

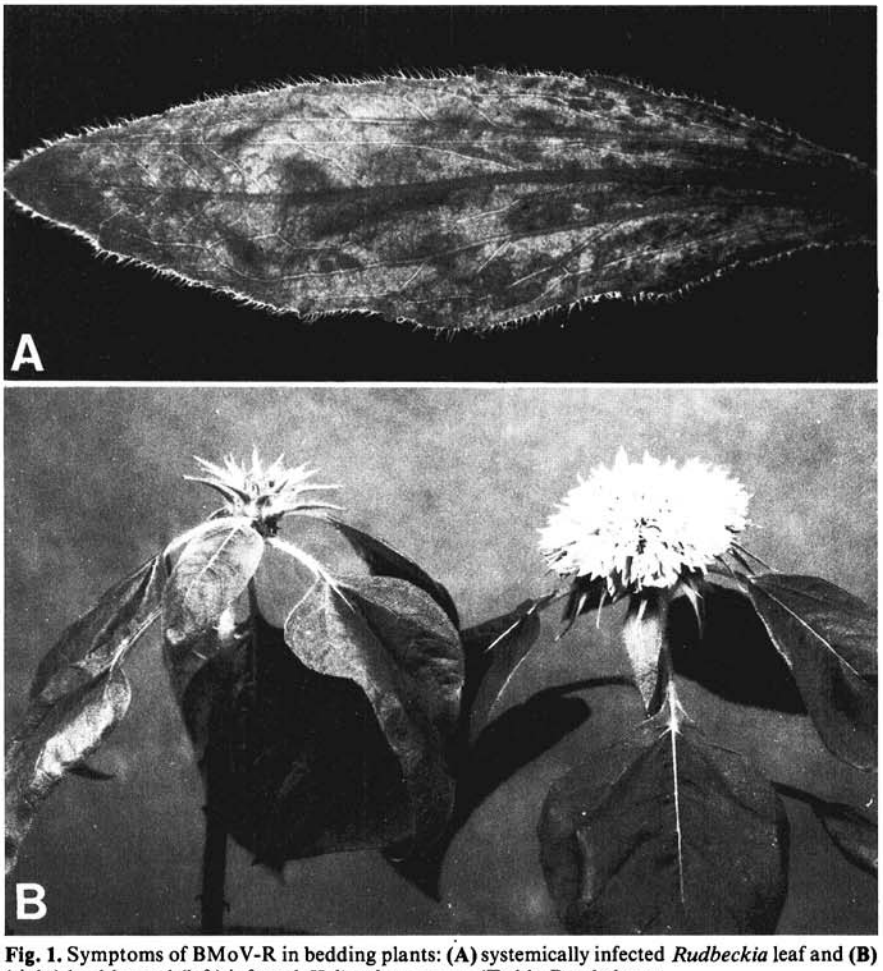

Fig. 1. Symptoms of BMoV-R in bedding plants: **(A)** systemically infected *Rudbeckia* leaf and **(B)** (right) healthy and (left) infected *Helianthus annuus* 'Teddy Bear' plants.

Light microscopy of stained epidermal tissues from BMoV-R infected *R. hirta hybridum, Z. elegans, and N.* × *edwardsonii* plants revealed cytoplasmic inclusions typical of potyviruses (5). Negatively stained leaf extracts from many of the inoculated bedding plants, *B. bipinnata* and *N.* × *edwardsonii* contained flexuous-rod particles. Also, striated laminated aggregate inclusions, somewhat rectangular in outline and resembling those described previously for BMoV (3), were detected in leaf extracts of BMoV-R-infected *N.* × *edwardsonii*.

In double radial immunodiffusion tests, BMoV-R reacted identically against either BMoV-E (10) or BMoV-F (11) antisera (Fig. 2). Homologous reactions did not spur over those with BMoV-R-infected leaf extracts of *Rudbeckia, N.* × *edwardsonii*, or other hosts (Table 1), and no reactions were seen when normal serum or leaf extracts of healthy plants were used. Extracts from 10 of 14 *Ageratum* plants collected from a bed in Alachua County, FL, and, 16 of 50 *Ageratum* plants collected from three locations in Orange County, FL, in December 1982 reacted identically against BMoV-E and BMoV-F antisera. Likewise, in July–August 1983, single specimens of *Centantherum intermedium* from Brevard County, FL, and *Z. elegans* from Alachua and Orange counties reacted with BMoV antiserum.

DISCUSSION

The virus infecting *Rudbeckia* was identified as BMoV on the basis of 1) characteristic symptoms in *N.* × *edwardsonii, Z. elegans, Helianthus annuus*, and *Chenopodium amaranticolor* (9), 2) light and electron microscopic observation of cylindrical inclusions closely resembling those noted for BMoV (3), and 3) reactions of identity noted in serological tests against BMoV-E and BMoV-F antisera. The virus collected from *A. conyzoides* and *Z. elegans* in Alachua and Orange counties was identified as BMoV on the basis of serological studies against BMoV-E and BMoV-F antisera.

This is the first report of natural infections of *R. hirta hybrida, Z. elegans*, and *A. conyzoides* by BMoV. This report also describes BMoV susceptibilty in 10 new species. One of these is *Verbena hybrida*, the first of the family Verbenaceae known to be susceptible to BMoV.

Bidens mottle virus was originally described in Florida in 1968 (3) and has since been found occurring naturally in

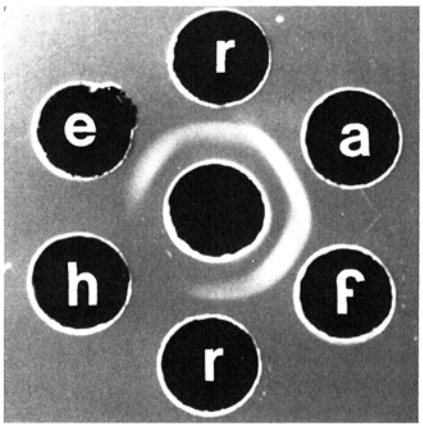

Fig. 2. Serological evidence for the susceptibility of certain bedding plants to BMoV. The center well contained antiserum to BMoV-F and the peripheral wells contained leaf extracts of the following: r = *Nicotiana* × *edwardsonii* manually inoculated with BMoV-R, a = *Ageratum* manually inoculated with BMoV-R, f = *Ageratum* field sample, *h* = healthy *Ageratum*, and e = *N. edwardsonii* manually inoculated with BMoV-E.

Florida as a pathogen of lettuce and endive (10), blue lupine (6), and the foliage ornamental *Fittonia* (11). BMoV also infects lettuce and endive in New York (R. Provvidenti, *personal communication*). The prevalence of this virus in Florida appears to reflect its relatively wide host range, which includes at least eight plant families, and its high incidence in common weeds such as *Bidens pilosa* and *Lepidium virginicum* (3). Thus, it is not surprising to find natural infections of this virus in bedding plants. In bedding plants, BMoV is most likely to be troublesome in such perennial plants as *R. hirta hybrida*, which can serve as a reservoir of inoculum for virus-free seedlings. Because this virus can cause debilitating symptoms in such bedding plants as *Calendula, Rudbeckia, Ageratum,* and *Helianthus*, it should be considered a potentially troublesome pathogen, particularly in Florida. Special care should be taken to avoid perpetuating suscepts of this virus, such as *R. hirta hybrida*, and propagating seedlings in areas near such weeds as *B. pilosa*, which might be infected with BMoV.

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