Occurrence of Southern Bean Mosaic Virus in Central Brazil

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ABSTRACT

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Bean plants infected with southern bean mosaic virus were found for the first time in central Brazil. Identification of the virus was based on host range, in vitro properties, electron microscopy, and serology. PI 313310 and Preto 153 were found to be good local lesion hosts for the virus.

Although many viruses are known to attack beans (*Phaseolus vulgaris* L.) in Brazil (1), bean "mosaico-em-desenho" virus (BMDV) and cowpea severe mosaic virus (CPSMV) are the only mechanically transmissible, isometric viruses known to infect this crop in central Brazil (2,7). During a survey of bean diseases in central Brazil in 1979 and 1980, an isometric virus that induced host reactions different from those of BMDV and CPSMV was isolated from several bean samples.

This paper reports isolation and properties of the virus, which was identified as southern bean mosaic virus (SBMV). Brief reports have been published previously (3,4).

MATERIALS AND METHODS

Samples of bean plants with virus symptoms were collected from an experimental plot at the Experimental Station of the University of Brasília and from several commercial plantings in Núcleo Rural do Rio Preto in the Federal District, Brazil, in 1979 and 1980, respectively. Extracts from triturated leaves were mechanically inoculated onto *P. vulgaris* 'Manteiga,' 'Rico 23,' and 'Jalo' plants. For host range and in vitro properties, other legume and nonlegume plants were also inoculated mechanically as previously reported (10), except that Celite was substituted for 400-mesh Carborundum.

The virus was maintained and multiplied on the systemically susceptible host Rico 23, whereas line PI 313310 and Preto 153 were used as local lesion hosts for virus assay. Virus purification, antiserum production, and serological tests were performed by the procedures described for CPSMV (8). An extinction coefficient of 5.85 mg/ml per centimeter at 260 nm (11) was used to estimate the concentration of purified virus preparations. Virus particles in leaf dips were detected as previously described (6).

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RESULTS

Isolation of the virus. In 1979, two samples of Rico 23 dry beans showing mild mosaic symptoms were collected at the Experimental Station. In 1980, samples of snap beans showing vein-clearing and mosaic symptoms were also collected from the commercial plantings. Leaf dip preparations from all these samples revealed isometric virus particles ca 28 nm in diameter. However, reactions on Rico 23, Manteiga, and Jalo beans were different from those induced by BMDV and CPSMV. BMDV causes local chlorotic lesions followed by top necrosis on Rico 23 or by mosaic on Manteiga and Jalo (3), whereas CPSMV induces only small local necrotic lesions on Rico 23 but local and systemic chlorotic spots in the other two cultivars (2). SBMV induced only mosaic in these cultivars. Also, crude sap of these plants gave negative results against BMDV and CPSMV antisera (8,9) in double-diffusion tests. Single-lesion isolates from PI 313310 and Preto 153 were used for further studies.

Host reactions. The virus infected only legume plants, inducing the following types of reactions: a) severe mosaic, stunt, leaf and pod deformation in Jalo bean and bean introduction lines UFV-694, 737, and 1208; b) severe mosaic in Cubano bean and bean introduction lines UFV-692, 693, 700, 705, 804, 897, 1223, and 1447; c) mild mosaic in line 6474 (*P. coccineus*) and in Alabama, Cuva 168-N, Manteiga, Manteigão Fosco, Porrillo, and Rico 23 beans and bean introduction lines UFV-679, 701, 708, 709, 710, 750, 751, 784, 896, 1208, 1454, and 1467; d) necrotic local lesions in Preto 153 and Pinto 111 beans, lines PI 313315 and PI 313310 (*P. coccineus*) (Fig. 1), line 201298 (*P. lunatus*), and cross line *P. vulgaris* × *P. coccineus* 3225; e) latent infection in Carioca, Honduras, Pintado, and Small White beans, bean lines PI 313234 and UFV-736, *P. coccineus* 'Amarelo Grande' and 'Preto,' and soybean (*Glycine max* (L.) Merrill) IAC-2.

The following legumes were immune to the virus: Bountiful, Caballero, Canário, Fígado de Galinha, Preto 143, Rapesão, and UFV-685 beans; cowpea (*Vigna unguiculata* (L.) Walp) Pitiúba and Seridó; *P. lunatus* PI 313313; and *P. coccineus* 'Branco' and 'Amarelo.' The virus also did not infect *Chenopodium amaranticolor* Coste & Reyn., *C. murale* L., *C. quinoa* Willd., *Euphorbia prunifolia* Jacq., *Gomphrena globosa* L., *Capsicum annum* L. 'Casca Dura,' *Lactuca sativa* L. 'Brasil 202,' *Lycopersicon esculentum* Mill. 'Santa Cruz,' *Nicotiana tabacum* L. 'TNN,' *N. rustica* L., and *N. glutinosa* L.

In vitro properties. The virus was inactivated by heating the juice of infected bean leaves for 10 min at 95 C but not at 90 C. The dilution endpoint was between 10^{-5} and 10^{-6}.

Serology. Crude sap from virus-infected plants formed a well-defined precipitin line in agar plates with an antiserum to SBMV. Crude sap did not react with antisera to CPSMV or BMDV or to bean pod mottle, bean rugose, bean yellow stipple, bean mild mosaic, peanut stunt, quail pea mosaic, blackgram mosaic, or cowpea mosaic viruses.

Virus purification and antiserum

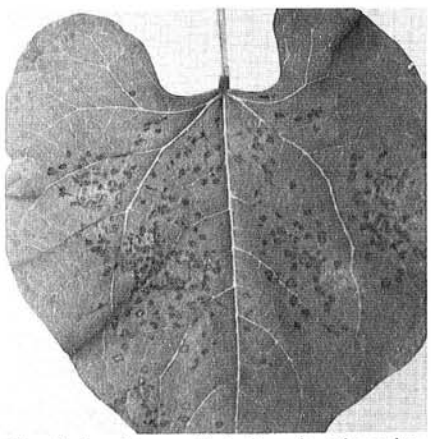

Fig. 1. Local necrotic lesions in *Phaseolus coccineus* PI 313310 15 days after inoculation with the Brazilian isolate of southern bean mosaic virus.

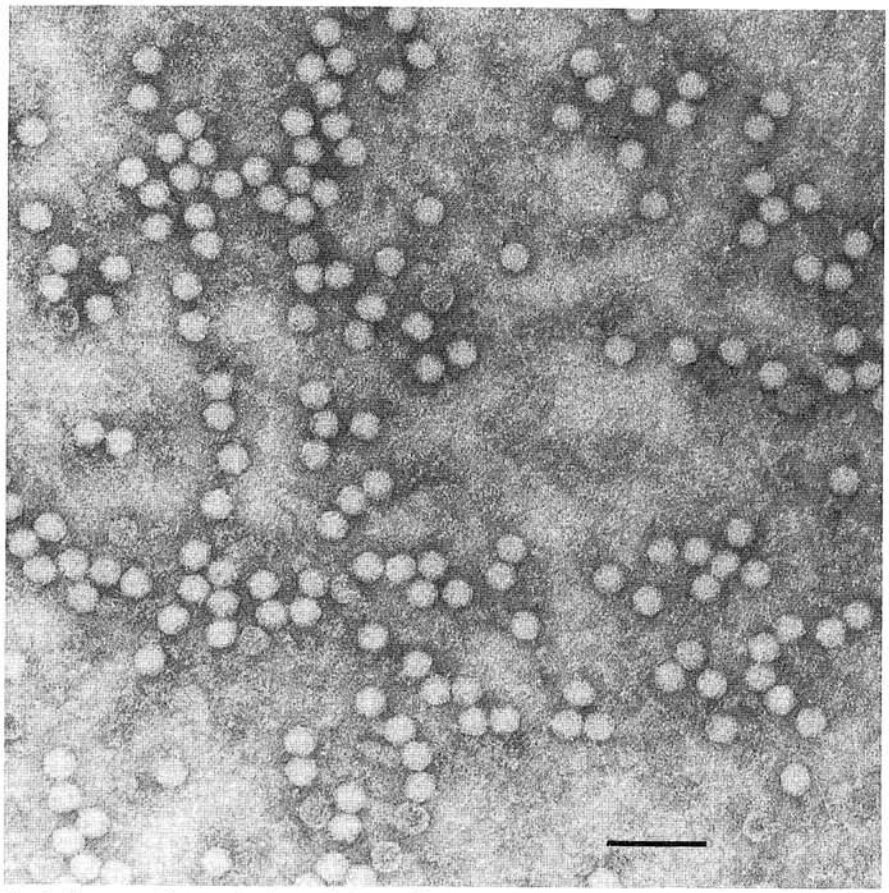

Fig. 2. Electron micrograph of a partially purified preparation of the Brazilian isolate of southern bean mosaic virus negatively stained with sodium silicotungstate. Bar = 0.1 μm.

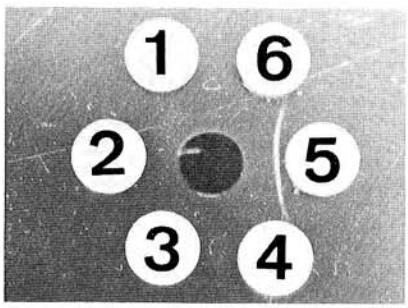

Fig. 3. Immunodiffusion pattern of the Brazilian isolate of southern bean mosaic virus. A 1:16 dilution of an antiserum to the Brazilian isolate was added to the central well, and the crude sap of six bean samples was placed in the peripheral wells. Note the well-defined, curved band between the antiserum and well 5, showing that only this sample was infected.

preparation. The virus was easily purified from leaves of systemically infected Rico 23 bean plants. Purified preparations contained numerous isometric particles ca 28 nm in diameter (Fig. 2). In sucrose density gradients, the virus sedimented as a single opalescent band 2.25 cm below the meniscus. The ultraviolet spectrum of purified preparations was typical of nucleo-protein with maximum and minimum absorptions at 260 nm and 243 nm, respectively, and $A_{260/280} = 1.54$. The yield was 120 mg of virus per kilogram of fresh leaf tissue.

The virus was highly immunogenic. An antiserum taken 18 days after the first injection had a titer of 1/512 in agar gel double-diffusion tests. A 1/16 or 1/32 dilution of this antiserum worked well to detect the virus in bean juice, forming a distinct, curved band (Fig. 3) in agar after overnight incubation in a moist chamber. The antiserum did not react with healthy bean juice.

DISCUSSION

Based on host range, particle morphology, thermal inactivation point, and serological relationships, the virus was identified as SBMV (11).

According to Shepherd (11), SBMV occurs in warm temperate and tropical regions of the Americas and Africa, and also in France. In Brazil, Costa et al (1) suggested that BMDV could be a member of the SBMV group, although they did not demonstrate serological or other relationships to SBMV. Based on this supposition, Gamez (5) included Brazil as one of the four countries in the Americas, and the only country in South America, where SBMV had been reported to occur. BMDV recently was shown to be a strain of bean rugose mosaic virus (9). Therefore, this is the first report of the field occurrence of SBMV in Brazil and South America.

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