Phaseolus lathyroides as a Reservoir of Cowpea Mosaic Virus in Puerto Rico

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ABSTRACT

Phaseolus lathyroides is usually infected by cowpea mosaic virus (CMV) in fields of western Puerto Rico. Symptoms of infection by the virus are usually masked by symptoms of bean golden mosaic infections. CMV was detected in bean (*Phaseolus vulgaris*) fields where infected *P. lathyroides* plants were commonly found. Characteristics of the virus were similar to those reported by other investigators.

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Phaseolus lathyroides L. is a common weed in Puerto Rico. It often has symptoms of virus infection, most commonly a bright yellow mosaic which has been attributed to the golden mosaic virus (J. Bird, *personal communication*). Bean golden mosaic is a serious disease of beans (*Phaseolus vulgaris* L.) in El Salvador (2), and is also found in other tropical countries (1).

In our attempts to transmit mechanically the bean golden mosaic virus from infected *P. lathyroides* plants, we observed several cases of infection of bean cultivars. Symptoms of infection were quite different from those reported for this species (1). We suspected, therefore, that *P. lathyroides* harbored more than one virus, that symptoms of infection by the other virus (or viruses) were masked by symptoms caused by the bean golden mosaic virus in *P. lathyroides*, and that several bean cultivars were susceptible to mechanical inoculation by this masked virus(s). We report here our observations on the infection of several hosts by this masked virus, its characteristics, and the possible role of *P. lathyroides* as a reservoir of the virus in fields of western Puerto Rico.

MATERIALS AND METHODS.—We made a host range study by inoculating several possible hosts of the virus. Inoculations were made with infective leaf sap of field-infected *P. lathyroides* plants in the first inoculations, and later with infective leaf sap of some of the species that were found susceptible to

infection. We inoculated week-old seedlings of the test species by rubbing the infective sap onto their leaves, which had been previously dusted lightly with Carborundum powder.

The Ouchterlony method was used to make serological observations of infected sap with several isometric legume virus antisera. Antisera of the following viruses were tested at a 1:8 dilution: alfalfa mosaic virus, broad bean mottle virus, cowpea mosaic virus (Arkansas strain), cucumber mosaic virus, cowpea chlorotic mottle virus, peanut stunt virus (Western strain), Pelargonium leaf curl virus, tomato black ringspot virus, tobacco ringspot virus, and Tephrosia yellow vein virus. Healthy and diseased leaves of *P. lunatus* (Henderson Bush cultivar) were used as sources of leaf sap. Infected leaf sap of the Henderson Bush cultivar were also used for observations of the virus particles under the electron microscope.

A survey was made of field-infected *P. lathyroides* in several sectors of western Puerto Rico to determine the percentage of infected plants with bright yellow

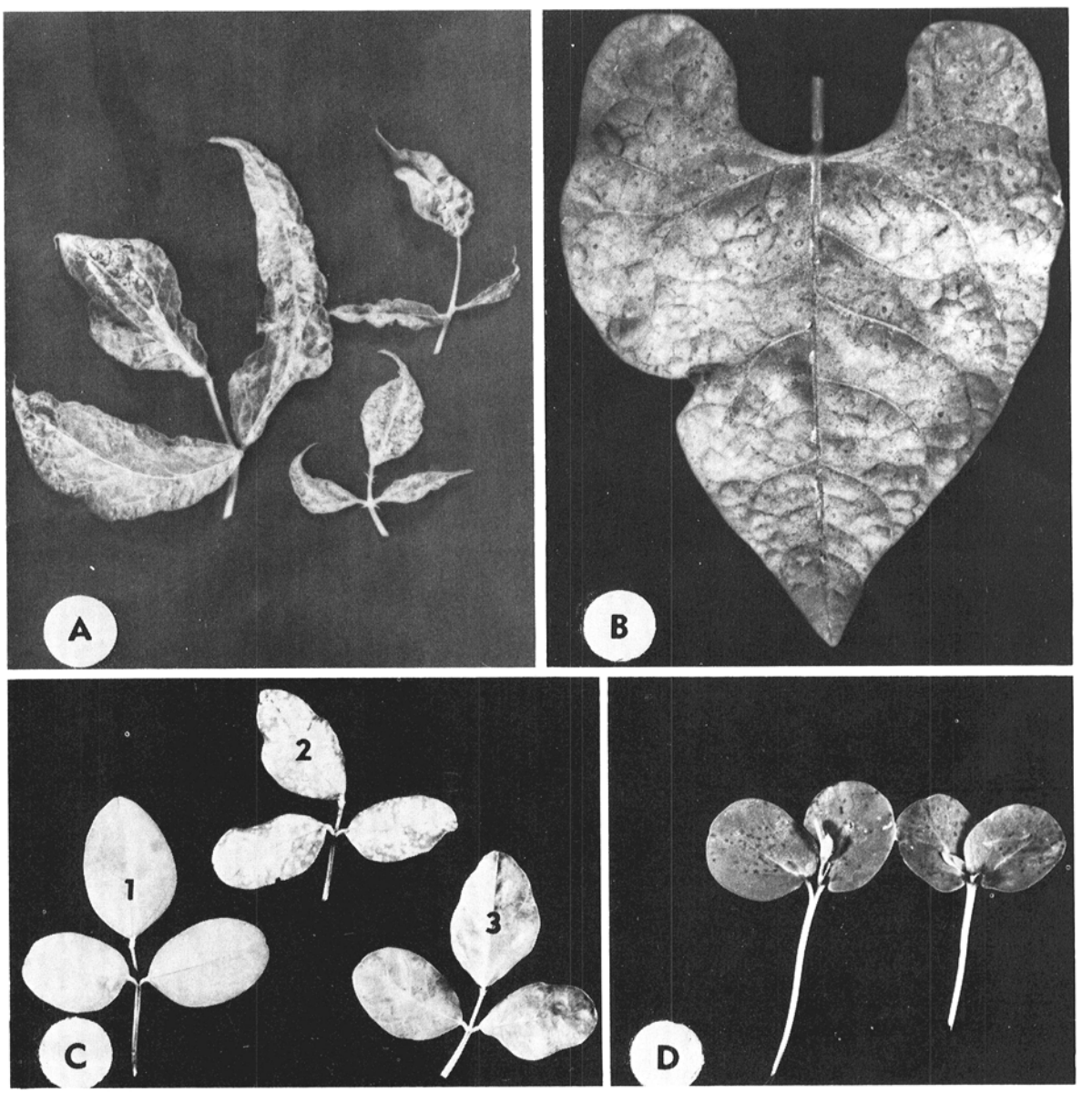

Fig. 1. Symptoms of infection by cowpea mosaic virus in selected species. **A)** Leaf malformation and mosaic in *Phaseolus vulgaris* cultivar Pure Gold. **B)** Local lesions in *P. lunatus* cultivar Henderson Bush. **C)** 1 = Light mosaic symptoms in field-infected *P. lathyroides;* 2 = golden mosaic symptoms in field-infected *P. lathyroides*; 3 = cowpea mosaic virus infection in mechanically infected *P. lathyroides.* **D)** Cowpea mosaic virus infections in mechanically infected *Cassia tora.*

mosaic symptoms which had the masked virus. The areas surveyed were fields in Aguada, Isabela, Rincon, the Las Mesas area of Mayaguez, and fields at the Federal Experiment Station in Mayaguez. Leaf samples were taken of each plant separately and indexed for infectivity on bean cultivars Black Valentine and Pure Gold as well as seedlings of *P. lathyroides*.

Field plantings of bean introductions in the Isabela and Mayaguez areas which had also many infected *P. lathyroides* plants were surveyed for infection by the virus. Leaf samples were taken at random from the fields and used to inoculate *P. lunatus* (cultivar Henderson Bush) seedlings in the greenhouse. Those plants that became infected and showed the typical red local lesions were used as inoculum sources for mechanical inoculation to Pure Gold, Black Valentine, *Cassia tora*, and *P. lathyroides* seedlings. This allowed a comparison of symptoms from this inoculum to others in different experiments.

A number of field-infected *P. lathyroides* plants showed also a dull green mosaic and light corrugations on their leaves. These symptoms could be seen in the same plants which had leaves with bright yellow mosaic symptoms. A test was made, therefore, to determine whether the dull green mosaic symptoms and leaf corrugations were caused by the mechanically transmitted virus which we had shown to infect *P. vulgaris*. Field-infected *P. lathyroides* with bright yellow and dull green mosaic with corrugated leaves were used to inoculate *P. lunatus* (cultivar Henderson Bush) seedlings in the usual manner.

Phaseolus lunatus (cultivar Henderson Bush) infected with the virus was used as an inoculum source to determine the dilution end point, thermal inactivation end point, inactivation time at room temperature (26 C), and inactivation time in leaves dried at 12 C. The same cultivar was used as an index host in these trials. Infected leaves of Black Valentine and Pure Gold were also used as inoculum sources to determine these properties. In this case, the index plants were healthy seedlings of both bean cultivars.

Seed transmission observations were made of field-infected plants known from previous tests to harbor the virus. Seeds of these plants were planted in the greenhouse and observed for symptoms of infection. At least 300 seedlings were observed for this purpose. Seeds of *Phaseolus lunatus* (cultivar Henderson Bush) and *P. lathyroides* plants which were mechanically infected were also tested for their transmission of the virus.

RESULTS.—*Infection of P. vulgaris cultivars with infected sap of field-infected P. lathyroides.*—Of the 20 bean cultivars tested with this inoculum source, only Black Valentine and Pure Gold became infected. Diffuse yellow spots developed on the inoculated leaves of both cultivars about 10 days after inoculation. Five days later, the inoculated plants showed malformed leaves, vein-clearing, mosaic, stunting, and shoot necrosis.

Host range, physical properties, and seed transmissions.—The host range of the mechanically transmitted virus was similar to that reported for cowpea mosaic virus (3, 4). The virus particles were isometric (ca. 28 nm diam), and infective leaf sap reacted only with cowpea mosaic virus antiserum. The virus was inactivated between 70 and 71 C in unamended leaf sap. Its dilution end point was between 10^{-4} and 10^{-5}. Infectivity was retained for 410 hr at 25 to 28 C. Desiccated leaves remained infective for at least 8 weeks at 12 C. No evidence of seed transmission was obtained.

Infection of P. lathyroides and P. vulgaris in the field.—*P. lathyroides* plants with golden mosaic symptoms from the areas surveyed were infected with the masked virus. When leaves with golden mosaic symptoms were used as inoculum source in some areas, 100% were shown to be infected with the masked virus. Only 20% of the plants with the dull green mosaic virus were found to be sources of it.

Bean introductions were also infected in fields when infected *P. lathyroides* was widespread, but to a lesser extent (10% of the samples taken). Some of these introductions (P.I.'s 30978, C-17791-C, and 309750) showed no outward symptoms of infection when sampled.

DISCUSSION.—The physical, serological, and host range characteristics of the virus were similar to those reported by other investigators for cowpea mosaic virus (3, 4). *Phaseolus lathyroides* usually harbors both the bean golden mosaic and cowpea mosaic viruses, is widespread in most fields in western Puerto Rico, and probably is a reservoir for infections to beans and cowpeas in field plantings.

P. lunatus 'Henderson Bush' is a useful test plant, as it shows distinct local lesions within a very short time after inoculation. *Cassia tora, Chenopodium amaranticolor, P. lathyroides*, and bean cultivars Pure Gold and Tendercrop are also useful species to differentiate the virus from others which are infective to *P. vulgaris* and other legumes (Fig. 1).

Source of inoculum seemed to be important in some inoculations. Leaves of field-infected *P. lathyroides* usually were a good source in inoculation to *P. lunatus, P. lathyroides*, or bean cultivars Pure Gold and Black Valentine. It was usually not a good source in inoculation to other bean cultivars. Leaves of infected *P. lunatus* were best as inoculum sources to test the susceptibility of these bean cultivars to the virus.

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