PEANUT MOTTLE VIRUS ISOLATED FROM SOYBEAN IN THAILAND

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Abstract

A virus isolated from soybean showing mosaic and stunting symptoms collected at Nakhorn Sawan, Northern Thailand in 1979, was identified as peanut mottle virus based on host range, symptomatology, aphid transmissibility, particle morphology, serological relationships, etc. The virus infected 15 plant species belonging to five families including systemic infection in peanut, pea, broad bean, etc., and also produced local lesions on inoculated leaves of Top Crop bean. The virus consisted of filamentous flexuous particles about 780 nm in length, and the thermal inactivation point, dilution end point, and longevity in vitro of the virus were 55 - 60 °C for 10 min, 10^{-3}-10^{-4}, and 7-14 days at 20°C, respectively. In ultrathin sections, pinwheel and bundle type inclusion bodies were observed in the cytoplasm. The virus reacted positively with antiserum to the Japanese isolate of peanut mottle virus.

1. Introduction

During the survey of virus diseases of soybean, soybean plant showing mosaic and stunting symptoms was collected in Northern Thailand in 1979. A virus isolated from this plant consisted of filamentous flexuous particles and infectious to peanut plant.

This paper describes some properties of the virus that was identified as peanut mottle virus.

2. Materials and methods

The virus used mainly in this experiment was isolated mechanically from soybean plants showing mosaic and stunting symptoms that were collected at Nakhorn Sawan in 1979. The virus was propagated from one local lesion on inoculated leaves of Top Crop bean and was maintained on Kintoki bean.

All the test plants were grown in a glasshouse. Mechanical inoculation was carried out by rubbing the surface of leaves dusted previously with Carborundum (600 mesh) with a cotton swab soaked in inoculum. Inoculum was prepared by grinding the leaves with 0.05 M phosphate buffer containing 0.01 M diethyldithiocarbamate and 1 mM L-cysteine.

Host range tests were carried out by mechanical inoculation to 27 plant species belonging to 9 families. The inoculum was prepared from infected Kintoki bean leaves.

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The infection of symptomless plants was confirmed by back inoculation to Kintoki bean, using sap extracted from inoculated leaves about 7 days after inoculation and from newly emerged leaves about 28 days after inoculation. All plant species were tested at least twice in different seasons.

Aphid transmission tests from diseased soybean to healthy soybeans were performed in using *Aphis glycines*. Aphids were allowed to fast for 2 hr before an acquisition access of 10 min. After the acquisition access, groups of ten aphids were transferred to test plants for the first inoculation access of 2 hr, then again transferred to a new test plant for a second inoculation access of 24 hr. These aphids were removed by spraying with insecticides.

Seed transmissibility was tested on progenies from seeds collected from infected soybean plants grown in glasshouse.

Stability in crude sap was determined based on the thermal inactivation point (TIP), dilution end point (DEP) and longevity in vitro (LIV) of the virus. Top Crop bean plants were used as assay plants.

For electron microscopy, small pieces of diseased soybean leaves were cut in a few drops of 2% potassium phophotungstate, pH 6.5, and a small amount of this preparation was dropped onto carbon-coated Formvar grids. Ultrathin sections were prepared from small pieces of diseased soybean leaves fixed in 3% glutaraldehyde in 0.1 M sodium phosphate buffer, pH 7.0, and post-fixed in 1% osmium tetroxide in the same buffer. Then, after dehydration in alcohol series, the materials were embedded in Spurr resin. Ultrathin sections were cut by using glass knives and stained by uranyl acetate and lead citrate. These preparations were examined under a Hitachi Model H-500 or H-300 electron microscope.

Serological relationships of the virus with peanut mottle virus from Japan were analyzed in ring test and immune electron microscopy.

3. Results

1) Host range and symptoms

The virus infected mechanically 15 plant species in 5 families among the 27 species in 9 families (Table 1). A few species belonging to the Leguminosae were infected systemically and showed symptoms.

Arachis hypogaea showed mild mottle symptoms systemically.

Glycine max showed distinct mosaic symptoms.

Phaseolus vulgaris 'Kintoki' was very susceptible, and showed chlorotic local lesions on inoculated leaves and vein-clearing and mosaic symptoms in newly emerged leaves.

Pisum sativum and *Vicia faba* showed systemically mosaic or chlorotic spot symptoms.

Phaseolus angularis, Vigna mungo, V. unguiculata, Nicotiana clevelandii, Petunia hybrida, and *Sesamum indicum* did not show any symptoms. However, back inoculation to Kintoki bean revealed that these plant species were infected with the virus systemically without symptoms.

Phaseolus vulgaris 'Top Crop' showed necrotic local lesions on inoculated leaves (Fig. 1), but was not infected systemically.

Chenopodium quinoa, Gomphrena globosa, Phaseolus vulgaris 'Yamashiro Kurosando', *Vigna radiata* and *Nicotiana tabacum* did not exhibit any symptoms. However, back inoculation to Kintoki bean revealed that inoculated leaves of these

Table 1. Host range of peanut mottle virus isolated from soybean

Plant	Symptom	
	IL	NIL
Chenopodium amaranticolor	—	—
C. quinoa	1	—
Spinacia oleracea	—	—
Gomphrena globosa	1	—
Tetragonia expansa	—	—
Brassica rapa	—	—
Arachis hypogaea	1	Mo
Glycine max	1	M
Phaseolus angularis	1	s
Phaseolus vulgaris 'Kintoki'	L	Mal, M, N
" 'Top Crop'	L	—
" 'Yamashiro Kurosando'	1	—
Pisum sativum	1	M
Triforium pratense	—	—
T. repens	—	—
Vicia faba	1	CS
Vigna mungo	1	s
V. rediata	1	—
V. sesquipedalis	—	—
V. unguiculata	1	s
Datura stramonium	—	—
Lycopersicon esculentum	—	—
Nicotiana clevelandii	1	s
N. glutinosa	—	—
N. tabacum 'Bright Yellow'	1	—
Petunia hybrida	1	s
Sesamum indicum	1	s
Cucumis sativus	—	—
Zinnia elegans	—	—

IL: inoculated leaves, NIL: non-inoculated leaves, Mo: mottle, M: mosaic, Mal: malformation, N: necrosis, CS: chlorotic spot, L: local lesion, 1 and s: symptomless infection on inoculated or non-inoculated leaves, respectively, —: no infection.

plants were infected without showing symptoms.

The virus did not infect 12 plant species.

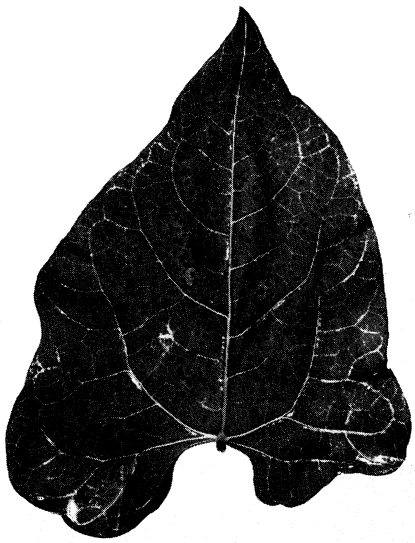

Fig. 1. Local lesions on inoculated leaf of Top Crop bean caused by peanut mottle virus.

2) Transmission

Aphid transmission. Aphid, *Aphis glycines,* transmitted efficiently the virus in the first inoculation access (8/10), but not in the second inoculation access (0/6). This result suggests that the virus was transmitted by aphids in a non-persistent manner.

Seed transmission. The virus was transmitted through seeds of soybean at a low percentage (1/452).

3) Stability in crude sap

The virus showed infectivity after sap was heated for 10 min at 55°C, but not at 60°C, diluted to 10^{-3}, but not 10^{-4}, and stored for 7 days at 20°C, but not 14 days.

4) Electron microscopy

Juice of infected soybean leaves contained filamentous flexuous particles about 780 nm in length.

In ultrathin sections, inclusions of pinwheel or bundle types were observed in the cytoplasm of infected soybean cells (Fig. 2).

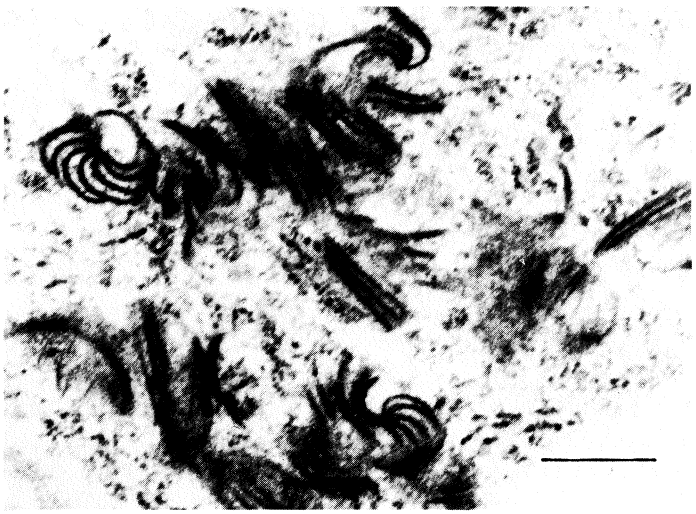

Fig. 2. Inclusion bodies in soybean cell infected with peanut mottle virus. Bar = 200 nm.

5) Serology

The virus reacted positively with antiserum to peanut mottle virus from Japan (1) (homologous titer : 1:256) diluted up to 1 : 256 in ring tests.

In immune electron microscopy tests, the virus particles were decorated completely with antibody, suggesting that these viruses have close serological relationships.

4. Discussion

The virus isolated from soybean showing mosaic and stunting symptoms was identified as peanut mottle virus (PnMV) based on host range, symptoms, transmission, particle morphology, serological relationships, etc.

PnMV was detected from only one sample of soybean, and from several samples of peanut.

The occurrence of PnMV on soybean had been reported (2) already, and occasional outbreaks occurred causing a considerable loss of yield of soybean.

Literature cited

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