Cowpea mosaic virus from Vegetable Soybeans in Korea

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Ninety samples showing mosaic symptoms on soybean (*Glycine max*) cv. Sukryangputkon were collected from the Cheongsonggun area, Kyungbuk province in Korea. Initially, DAS-ELISA was conducted for detection of *Soybean mosaic virus* (SMV). Negative samples were chosen at random and mechanically inoculated on soybean cv. Buffalo, which reported not to produce mosaic symptoms when mechanically inoculated with SMV. An isolate of SMV, designated as B-1, from Buffalo showing mosaic and mottle symptoms was used for identification and biological characterization of the causal virus. The purified B-1 isolate had spherical particles of approximately 24 nm. It positively reacted with the antiserum against *Cowpea mosaic virus* (CPMV) but not with *Cucumber mosaic virus* (CMV) and SMV antiserum. CPMV was newly isolated from soybean and had been characterized by host range and by serological and electron microscopic methods. Results of this study suggest that CPMV is the possible cause of mosaic disease in vegetable soybean and that based on symptomatology, a difference between the typical mosaic and rugose symptoms caused by SMV and CPMV was observed. This is first report of CPMV from soybean in Korea.

Keywords: *Cowpea mosaic virus*, identification, serology, soybean

Materials and Methods

Virus source and isolation of causal virus. Ninety samples showing mosaic symptoms on soybean cv. Sukryangputkon were collected from the Cheongsonggun area, Kyungbuk province in Korea. DAS-ELISA was conducted for detection of SMV, and negative samples were selected. Negative samples were chosen at random and mechanically inoculated on plants of soybean cv. Buffalo, previously reported not to produce mosaic symptoms when inoculated with SMV (Cho, 1978).

An isolate, designated B-1, from Buffalo showing mosaic and mottle symptoms were separated by successive single local lesion methods and used for identification and biological characterization of the causal virus (Fig. 1). The soybean cv. Buffalo was used to increase virus inoculum and to exclude possible mixed infec-

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Several cases of severe mosaic disease infestation were observed on vegetable soybeans (*Glycine max*) in Cheongsong area, Kyungbuk province in Korea in 2000. *Soybean mosaic virus* (SMV) was considered as the possible cause of the mosaic disease. SMV has been known as a prevalent pathogen and is widely spread in Korea. Meanwhile, *Cowpea mosaic virus* (CPMV) was newly isolated from soybean and has been characterized by host range and by serological and electron microscopic methods. This paper reports that CPMV is the possible cause of mosaic disease in vegetable soybean and that based on symptomatology, a difference between the typical mosaic and rugose symptoms caused by SMV and CPMV was observed.

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Fig. 1. Symptoms of Buffalo (soybean cultivar) inoculated with ELISA negative samples. a: mosaic symptom of noninoculated upper leaf, b: more severe symptom of new emerging leaves.


Table 1. Reactions of isolate B-1, *Cowpea mosaic virus*, *Soybean stunt virus* and *Alfalfa mosaic virus* on different indicator plants

<table>
<thead>
<tr>
<th>Indicator plant</th>
<th>Symptoms of B-1 isolate and other related legume virus*</th>
<th>AMV*</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Chenopodium amaranticolor</em></td>
<td>L/M, L/M</td>
<td>L/M</td>
</tr>
<tr>
<td><em>Gomphrena globosa</em></td>
<td>-/-, -/-</td>
<td>L/YS, L/M</td>
</tr>
<tr>
<td><em>Cucumis sativus</em></td>
<td>-/-, -/-</td>
<td>-/-</td>
</tr>
<tr>
<td><em>Datura stramonium</em></td>
<td>-/-, -/-</td>
<td>-/-</td>
</tr>
<tr>
<td><em>Pisum sativum</em></td>
<td>-/-, Mo, Mo, Mo, Mo, L/M, Mo</td>
<td>L/M</td>
</tr>
<tr>
<td><em>Phaseolus vulgaris</em> cv. Top Crop</td>
<td>-/-, Mo, Mo, Mo</td>
<td>L/-</td>
</tr>
<tr>
<td><em>Phaseolus vulgaris</em> cv. Scotia</td>
<td>-/-, Mo, Mo, L/M</td>
<td>L/-</td>
</tr>
<tr>
<td><em>Vigna unguiculata</em> cv. Blackeye</td>
<td>L/M, L/M, Mo, Mo</td>
<td>L/-</td>
</tr>
<tr>
<td><em>Vigna unguiculata</em> cv. Early Ramshorn Blackeye</td>
<td>L/M, L/M, Mo, Mo</td>
<td>L/-</td>
</tr>
<tr>
<td><em>Vigna unguiculata</em> cv. Chinese Red</td>
<td>L/M, Mo, Mo, Mo</td>
<td>L/-</td>
</tr>
<tr>
<td><em>Nicotiana glutinosa</em></td>
<td>-/-, Mo, Mo, L/M</td>
<td>L/M, Mo</td>
</tr>
</tbody>
</table>

*Symptoms on inoculated leaves/noninoculated upper leaves; –: symptomless, M: mosaic symptoms, L: local lesion, YS: yellowing spot, Mo: mottling symptoms, I: latent infection.*

*Cowpea mosaic virus (CPMV), Soybean stunt virus (SstV) and Alfalfa mosaic virus (AMV) were reported by Hampton et al. (1978).*

**Results and Discussion**

**Host reaction.** In the host range tests, *Chenopodium amaranticolor* inoculated with B-1 caused local lesions on the inoculated leaves and mosaic symptoms on the upper leaves (Table 1). Legume bean and cowpea cultivars produced mosaic symptoms similar to CPMV but different from those of *Soybean stunt virus*, a legume strain of CMV (Hanada and Tochihara, 1982) and *Alfalfa mosaic virus* (Hampton et al., 1978; Table 1). Soybean differentials resistant to mosaic disease producing necrosis to virulent SMV strain G7 produced mosaic symptoms (Table 2).

**Sero logical relation and morphology of the virus.** The positive results in agar double diffusion tests (Fig. 2) strongly suggested that the isolate B-1 is an isolate of...
Fig. 2. Serological reaction in agar gel double diffusion test (0.7% agarose gel in 0.02 M phosphate buffer, pH 7.0). Photograph: A: Soybean plant sap infected with SMV-G5, B: 3% SDS (control), C: tobacco plant sap infected with CMV, D: soybean plant sap infected with B-1 isolate, central well contain SMV antiserum (ATCC, PVAS-94). Photograph: a: tobacco plant sap infected with CMV, b: soybean plant sap infected with SMV-G5, c: soybean plant sap infected with B-1 isolate, d: healthy tobacco plant sap (control), central well contain CMV antiserum. Photograph: A: soybean plant sap infected with B-1 isolate, B: tobacco plant sap infected with CMV, C: soybean plant sap infected with SMV-G5, D: healthy soybean plant sap (control), central well contain CPMV antiserum.

Fig. 3. Ultra-violet absorption spectra of purified virus from B-1 isolate.

CPMV. The B-1 virus particles were isometric about 24 nm in diameter and associated with membrane structures in the cytoplasm (Figs. 3 and 4; Van der Scher and Groenewegen, 1971). In conclusion, results of this study suggest that the causal pathogen in the soybean was an isolate of CPMV based on serological and ultrastructural properties. This is the first report of CPMV from soybean in Korea.

Although beetles were reported as the vectors of CPMV, epidemiological aspects (Ross, 1968) of vegetable soybean mosaic disease caused by B-1 isolates, such as inoculum source, disease severity, and vector identification should be investigated to have a better knowledge of control measures for the virus.

Acknowledgment

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References


Puricifull, D. E. and Batchelor. 1977. Immunodiffusion tests with
Fig. 4. Accumulation of virus-like particles in buffalo leaves A: particles in the cytoplasm (×2500), B: membrane structures in the cytoplasm and particles in the vacuole (×8000), C: electron micrograph of purified virus from B-1 isolate (×60000). Virus particles were negative stained with 2% uranyl acetate.