# Peanut Stunt Virus in Crownvetch

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ABSTRACT

A virus isolated from crownvetch, *Coronilla varia,* was identified as a member of the cucumber mosaic virus group. The virus was determined to be identical to peanut stunt virus on the basis of host range, symptomatology, physical properties, serological reactions, and aphid transmissibility. Of 30 plant introductions of *C. varia,* inoculated with the virus, 16 were infected and showed definite symptoms, 10 showed very mild symptoms, and four were not infected. Also, *C. globosa, C. coronata, C. scorpioides,* and a *Coronilla* sp. were hosts for the virus. Widespread use of crownvetch along highways potentially can provide tremendous reservoirs of virus and insect vectors for transmission to susceptible agronomic hosts.

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The legume, crownvetch (*Coronilla varia* L.), is grown widely in Virginia and other states on highway slopes and as a grazing and forage crop. Few diseases have been reported on crownvetch. Ostazeski and Scott (9) were the first to report a virus from naturally infected crownvetch. They described a virus isolated from a space planting in Blacksburg, Virginia, and concluded that it was cucumber mosaic virus (CMV). In our study, we more precisely identified a virus isolated from the same site by comparing it with peanut stunt virus and tested the reaction of various *Coronilla* introductions and species to it.

MATERIALS AND METHODS.—The crownvetch virus (CVV) was first isolated from crownvetch (cultivar Emerald) leaflets ground in 0.01 M sodium phosphate (pH 7.0) and inoculated to primary leaves of cowpea [*Vigna unguiculata* (L.) Walp., 'Early Ramshorn']. For later isolations and routine transmissions, we rubbed crownvetch leaflets directly onto peanut (*Arachis hypogaea* L. 'Virginia 56R'). Sap inoculations were used in the host range study. All plants were dusted with (600-mesh) Carborundum before inoculation. Leaves were rinsed with tap water after sap inoculations, but were not rinsed after leaf-rub inoculations. Aphid transmission was tested as per Isakson (5), with the green peach aphid [*Myzus persicae* (Sulzer)].

The virus was purified from infected cowpea by the same methods used for peanut stunt virus (PSV) (10).

Serological relationships were tested by Ouchterlony gel diffusion tests, by use of 0.8% Ionagar No. 2 with 0.1% sodium azide, or by a modification of the density-gradient serology technique (4). Antisera used in the tests were anti-PSV, prepared by Groelke (4) to a Virginia isolate of PSV, and anti-CVV, kindly supplied by H. A. Scott, Fayetteville, Arkansas.

Seedlings of 38 plant introductions of crownvetch, obtained from the U.S. Department of Agriculture Regional Plant Introduction Station at Ames, Iowa, were transplanted into flats and maintained in a greenhouse throughout the experiment. Inoculations were made 2 weeks after transplanting by the leaf-rub method from peanut. Symptom expression was read at 3, 8, and 23 weeks after inoculation. At the time of the readings, we trimmed plants back to the crowns and took no precautions to prevent horizontal virus transmission. At 28 weeks after inoculation, single plants were indexed to single peanut plants, and results were recorded 4 weeks later.

RESULTS.—*Host range, symptomatology, and transmission.*—Symptoms appearing on naturally infected crownvetch plants consisted of a mild, chlorotic mottle and a slight distortion of the youngest leaflets. No stunting or reduction in vigor of the plants was noted. Symptoms induced by the virus in peanut were the same as those described by Miller and Troutman (6) for PSV. Other hosts in which the CVV induced symptoms similar or identical to those induced by PSV included cowpea, pea (*Pisum sativum* L. 'Little Marvel'), lima bean (*Phaseolus limensis* Macf. 'Fordhook'), bean (*P. vulgaris* L. 'Top Crop' and 'Red Kidney'), tobacco (*Nicotiana tabacum* L. 'Burley 21', 'Vesta 5', and 'Xanthi'), and goosefoot (*Chenopodium amaranticolor* Coste & Reyn.). The green peach aphid transmitted CVV in a stylet-borne manner from either crownvetch or peanut to seedling peanut, in which typical stunt symptoms developed within one week.

*Physical properties and serology.*—For partial purification of CVV, cowpea primary leaves were homogenized in 0.05 M sodium phosphate (pH 7.7) containing 0.2% sodium diethyldithiocarbamate, emulsified with an equal volume of chloroform:butanol (1:1, #v:v), and centrifuged. Virus preparations formed a single infectious zone in sucrose density-gradient columns, which cosedimented with PSV. Samples from the CVV zones contained uniform icosahedral particles, which were indistinguishable from PSV in the electron microscope.

Purified CVV and crude sap from infected cowpea reacted with antiserum to PSV in agar-gel diffusion tests. Cross-reactions with purified PSV and Scott's CVV antiserum, showed that the two viruses were serologically identical. Identity was shown also by complete removal of homologous and heterologous virus zones after incubation of the antigen-antibody mixtures and analysis by density-gradient centrifugation.

*Reaction of crownvetch introductions to PSV.*—The plant introductions were segregated on the basis of their response to CVV, and the results are shown in Table 1. Classifications were based on general appearance of the inoculated plants, and on recovery of virus to peanut at the end of the tests. Because no plants were left uninoculated in this test, symptoms alone were not used as a measure of the response. The four entries classed as resistant showed no mottling symptoms, and indexing to

TABLE 1. Response of *Coronilla* introductions to the crownvetch virus

| Class | Species | P.I. Number | | | |
|---|---|---|---|---|---|
| Resistant | *C. varia* | 228373 | 228411 | 229968 | 274041 |
| Moderately resistant | *C. varia* | 206487 | 210365 | 229627 | 230340 |
| | | 238142 | 278698 | 286203 | 308009 |
| | | 308980 | 325265 | | |
| Moderately susceptible | *C. varia* | 204871 | 251808 | 274040 | 325255 |
| | | 325257 | 325258 | 325259 | 325260 |
| | | 325261 | 325262 | 325263 | 325264 |
| | | 325266 | 325324 | 326369 | 340779 |
| | *C. globosa* | 283240 | | | |
| | *C. coronata* | 326366 | | | |
| | *Coronilla* sp. | 253435 | | | |
| Very susceptible | *C. scorpioides* | 186299 | 244311 | 287792 | 287793 |
| | | 287794 | | | |

TABLE 2. Average vigor score[a] of plant introductions according to virus reaction class at Blacksburg, Virginia

| Class | Vigor Score | | |
|---|---|---|---|
| | 1971 | 1972 | 1973 |
| Resistant | 3.42 | 3.1 | 2.2 |
| Moderately resistant | 3.34 | 2.7 | 2.0 |
| Moderately susceptible | 2.95 | 2.6 | 2.0 |
| Very susceptible | 3.41 | 4.4 | 2.4 |

[a]Scale of 1 = best to 5 = poorest.

peanut was negative. Ten moderately resistant entries showed very mild or no mottling, and indexing of some or all of the plants was positive. The remaining 16 *C. varia* entries were classed as moderately susceptible. These plants typically showed some mottling, and leaflets were often smaller, curled at the edges, and twisted on the petiole. However, little or no overall effect on vigor of the plant was noticed.

Symptoms on other species of *Coronilla* were more striking than on *C. varia*. *Coronilla globosa* Lam., a perennial introduction from Australia, was markedly yellowed and stunted. Few new branches were formed after infection. *Coronilla coronata* L. showed a well-defined, chlorotic mottle on some, but not all, of the leaflets on a plant. Also, leaflets were twisted on the petiole, so that they were no longer in a uniform alignment. The five introductions of *C. scorpioides* (L.) Koch were severely affected by the virus, showing a definite mottle soon after inoculation, severe stunting, and death before the final reading.

Mild mottling symptoms were induced by CVV on three common cultivars of crownvetch (Penngift, Emerald, and Chemung) and the virus could be recovered to peanut from each of these cultivars.

*Relationship of geographic origin to virus reaction.*—All but one of the resistant plant introductions was from Iran. The original source of the other resistant strain (274041) is unknown, but it also may have been from Iran. Moderately resistant strains included three from Iran, two each from Turkey and Czechoslovakia, and one each from Iowa, Rumania, and USSR. None of the moderately susceptible introductions came from Iran; 13 of the 19 came from USSR. Two came from Iowa, and one each from Turkey, Yugoslavia, and Australia. No *C. varia* strains were as susceptible as those of *C. scorpioides*.

*Relationship of vigor and resistance.*—Vigor of each of the plant introductions in the field was rated for three consecutive years in Blacksburg, on a scale from 1 (best) to 5 (poorest). The average rating of strains for each virus reaction class is shown in Table 2. No consistent relationship appeared, because very susceptible plants were vigorous as were resistant plants. Among the *C. varia* entries, however, the resistant entries averaged consistently more vigorous. Because these plots were not examined for the presence of virus, the interaction of PSV and vigor is not known.

DISCUSSION.—The virus isolated from crownvetch, CVV, is probably the same one that Ostazeski and Scott (9) described as CMV. Comparison of CVV with PSV shows them to be very similar, if not identical. PSV is a member of the CMV group, but differs enough from most CMV strains in biological, serological, and chemical properties that it is considered to be a distinct virus (1, 7, 11). Besides peanut, PSV has been isolated also from naturally infected bean (2, 8), tobacco (3), hoary-pea (*Tephrosia*) (13) and soybean (S.A. Tolin, *unpublished*). White clover has been considered to be the primary overwintering host (12). Crownvetch, another perennial host of PSV, may serve as a source of virus for epiphytotics. In this study, we did not try to determine the extent and distribution of PSV in crownvetch in Virginia.

Three commonly grown cultivars of crownvetch (Chemung, Emerald and Penngift) were susceptible and showed mild symptoms. All but four perennial plant introductions of crownvetch were susceptible. Crownvetch is used increasingly along roadsides and in pastures, and it possibly carries a "latent" infection with PSV. Therefore, epidemics could result if susceptible crops were grown near crownvetch plantings, and if aphid vector populations were high early in the growing season. The use of PSV-resistant plant introductions would be advisable in developing acceptable cultivars of crownvetch. PSV appears to be the most important of the common legume viruses to be considered in breeding programs, because crownvetch was not infected by alfalfa mosaic virus, bean pod mottle virus, soybean mosaic virus, or peanut mottle virus (Tolin, *unpublished*).

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