

Beetle Transmission of Cowpea Chlorotic Mottle Virus

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ABSTRACT

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Cowpea chlorotic mottle virus was transmitted by the bean leaf beetle, *Cerotoma trifurcata*, and by the spotted cucumber beetle, *Diabrotica undecimpunctata howardii*. Three strains of the virus were transmitted at low levels. Levels of transmission were highest when virus was acquired

from and transmitted to beans. Levels were much lower when the virus was either acquired from or transmitted to cowpeas. *Lespedeza cuneata*, which may be an overwintering host, was a poor source of virus for vectors.

Several viruses in the bromovirus group are transmitted by leaf-feeding beetles. Cowpea chlorotic mottle virus (CCMV) was transmitted by the chrysomelid species *Cerotoma trifurcata* (Forster) and *Diabrotica undecimpunctata howardii* (Barber) (6); the bean yellow stipple strain (BYSV) of CCMV by *C. ruficornis* Olivier and *D. balteata* LeConte (2,3); and broad bean mottle virus (BBMV) by *D. undecimpunctata howardii*, *Acalymma trivittatum* (Mannerheim), and *Colaspis brunnea* (F.) (7). Levels of transmission were moderate to low in all cases. Because CCMV is common in the southern United States and Central America (1), this study was undertaken to evaluate beetle transmission of CCMV in detail.

MATERIALS AND METHODS

Three serologically distinct strains of cowpea chlorotic mottle virus (CCMV, BYSV, and CCMV-A) (1) were tested for beetle transmission. Plants used for virus acquisition source or transmission tests included bean (*Phaseolus vulgaris* L. 'Pinto'), cowpea (*Vigna unguiculata* (L.) Walp. 'Monarch'), Florida beggarweed (*Desmodium tortuosum* (Sw.) DC), and *Lespedeza cuneata* (Dumont) G. Don. Plants were grown from seed in the greenhouse except for lespedeza, which was potted from field-grown plants.

The spotted cucumber beetle *D. undecimpunctata howardii*, the bean leaf beetle *C. trifurcata*, and the Mexican bean beetle *Epilachna varivestis* Mulsant were tested for ability to transmit the virus strains. The Mexican bean beetle and some bean leaf beetles were reared in the laboratory (4). The spotted cucumber beetle and some bean leaf beetles were field-collected. Laboratory-reared beetles were from colonies known to be free of plant virus contamination. Field collections were from soybean plantings without evident CCMV, and testing representative samples before and during trials revealed no beetles carrying CCMV.

For 1 or 2 days, beetles were given access to excised, infected leaf tissue in plastic cups. Beetles that fed during acquisition access were confined individually on test plants to test for transmission. Beetles were transferred to new test plants daily to determine length of virus retention. Acquisition and test feedings were at 28 C. Test plants were observed for virus symptoms for 3 wk. Transmission was verified by allowing plant sap to react with virus-specific

antisera, using the Ouchterlony gel diffusion technique. Virus in beetles that had fed on infected plants was tested by severing one metathoracic leg and collecting in a capillary tube the hemolymph that exuded. Hemolymph was diluted approximately 1:300 in 0.01 M phosphate buffer, pH 7.2, and inoculated to young Monarch cowpea plants.

RESULTS

Levels of transmission of three strains of CCMV by the bean leaf beetles did not differ. When bean was both the acquisition source and test plant, approximately 12% of beetles transmitted CCMV, BYSV, or CCMV-A. In trials using bean for both acquisition and transmission hosts, virus was transmitted to 19 of 117 plants (16.2%) by spotted cucumber beetles, compared with 47 of 376 (12.5%) by bean leaf beetles. Transmission of CCMV by the Mexican bean beetle occurred only occasionally.

Bean was a better host than cowpea for acquisition and transmission of CCMV by the bean leaf beetle. In all trials the rates of transmission were highest when bean was both the acquisition source and the test plant. Bean leaf beetles' level of transmission was approximately 12% when bean was both the acquisition and transmission host. This contrasted with transmission at 2% when cowpea was either the acquisition or test host. When beetles were fed on infected cowpea plants and transferred to bean plants, only three of 131 transmitted virus. When infected bean plants were the acquisition host and beetles were transferred to cowpea, only one of 75 transmitted virus.

During this study, naturally infected *L. cuneata* was found near field plantings of bean and cowpea containing some CCMV-infected plants. This plant and *D. tortuosum* were evaluated as virus acquisition hosts for the bean leaf beetle. Four of 48 beetles that fed on infected *D. tortuosum* transmitted virus to bean, but only four of 105 beetles that fed on *L. cuneata* transmitted virus.

Bean leaf beetles typically transmitted CCMV no longer than 3 days after virus acquisition. Occasionally beetles transmitted after 4 or 5 days, and one beetle transmitted virus on the seventh day after acquisition. This beetle had fed on 3 of the prior 6 days but had not transmitted CCMV. Only one beetle transmitted virus to more than one plant; this was on the first and third days after acquisition.

Hemolymph may be a reservoir for some beetle-transmitted viruses (5). Virus was detected in the hemolymph of two of 60 beetles fed on CCMV-infected plants.

DISCUSSION

Viruses in the bromovirus group are transmitted at low levels of efficiency by beetles (2,3,6,7), and these studies present no exception. Our results suggest that leaf feeding beetles transmit CCMV less efficiently than cowpea mosaic virus, bean pod mottle virus, southern bean mosaic virus or the cowpea strain of southern bean mosaic virus. Furthermore, consistent differences in ability to acquire and transmit virus from bean and cowpea were evident. The efficiency of transmission was very poor when cowpea was involved. No reason for the difference between these two hosts was apparent. The virus could be transmitted readily to either host by mechanical means. The beetles fed equally well on either bean or cowpea. Because levels of transmission were reduced when cowpea was either the acquisition or transmission host, concentration of virus during acquisition probably would not account for the difference.

A possible overwintering source of virus, *L. cuneata*, was not a good virus acquisition source. It could, however, serve as a virus source for overwintering beetles that emerge in the spring.

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