Beet leafhopper transmits virescence of periwinkle

George N. Oldfield ■ George H. Kaloostian ■ Harold D. Pierce ■ Andrew L. Granett ■ Edmond C. Calavan

Photo above shows healthy periwinkle on left and virescence-affected periwinkle on right; plant on right was exposed to beet leafhopper *(Circulifer tenellus)* that had previously fed on another virescence-affected periwinkle. Photo below shows periwinkle plant that developed virescence after exposure to infective beet leafhopper; note variation in severity of symptoms.

During studies of the relationship of various leafhopper species to the spread of stubborn disease of citrus, the periwinkle plant, *Vinca rosea*, was shown to be a host of *Spiroplasma citri*, the organism that causes stubborn disease. Also, one species of leafhopper, *Scaphytopius nitridus*, was shown to be capable of transmitting *S. citri* from citrus to periwinkle (see *California Agriculture*, February 1975). Once infected, the periwinkle plant developed abnormally small flowers and yellow foliage and then wilted and died within a month.

In view of this marked reaction to infection by *S. citri* and the apparent ease with which the organism was transmitted to periwinkle by *S. nitridus*, we began to use periwinkle in determining whether other species of leafhoppers collected in the field carried *S. citri*. We found that *Circulifer tenellus* collected at several locations in California carried *S. citri* and transmitted it to both citrus and periwinkle (see *California Agriculture*, June 1976).

Virescence developed

We subsequently observed that many of the periwinkle plants exposed to *C. tenellus* collected in the field at Riverside and Moreno, Riverside County, in early October 1974 developed virescence and rosetting. Virescence is a condition in which the flower petals are green rather than the normal color (pink, white, etc.). Rosetting is an abnormality characterized by shortening of the internode, or length of the stem between leaves.

We noticed the same symptoms on periwinkles exposed to *C. tenellus* collected later in October 1974 from the Mojave Desert near Victorville and Helendale, San Bernardino County. In all, 25 periwinkle plants exposed to *C. tenellus* collected in October 1974 developed virescence.

At first we suspected that the virescence might be a heretofore unrecognized symptom of infection of periwinkle by *S. citri*; however, when we attempted to isolate *S. citri* from each of the 25 plants showing virescence, we found that only one plant harbored this organism. That plant had first developed slight yellowing on a few leaves, which suggested infection by *S. citri*. Later it developed typical virescence, and the yellows disappeared. At that time, we could no longer isolate *S. citri* from it. All 25 plants showing virescence remain alive in the greenhouse two years after the appearance of virescence. Repeated attempts to isolate *S. citri* from each plant have been unsuccessful.

Not all periwinkle plants exposed to the 1974 field-collected *C. tenellus* developed virescence. Some of the plants that developed virescence exhibited symptoms on most or all branches soon after the first sign of abnormality; others retained normal flowers on some branches for several months before virescence and the accompanying rosetting finally developed throughout the plant.

Leafhopper as vector

On the basis of these observations, we suspected that the 1974 field-collected *C. tenellus* had transmitted to periwinkle some agent that caused virescence. The development of virescence in many species of plants has been attributed to infection by mycoplasma-like organisms, some of which are known to be transmitted by leafhoppers. For this reason, we suspected that the virescence in our periwinkle plants might be caused by an organism that had been transmitted by the field-collected C. *tenellus*. At the same time, *C. tenellus* is well known as the vector of the curly top virus of sugar beets and other plants, so we also needed to ascertain whether the curly top virus might have caused the virescence.

To verify that *C. tenellus* had transmitted an agent that caused virescence and to ascertain whether the agent was the curly top virus, we exposed groups of laboratory-reared *C. tenellus* known to be free of any plant pathogen to each of the periwinkles with virescence. After one week, we transferred each group to one healthy sugar beet for one day (a sufficient time for transmission of curly top virus); then we transferred each group to a healthy periwinkle plant and allowed the leafhoppers to feed until they died.

The leafhoppers acquired curly top virus from 9 of the 25 periwinkle plants with virescence and transmitted it to sugar beet. Six of these nine groups of leafhoppers subsequently transmitted the virescence agent to periwinkle plants. Two groups that did not transmit curly top to sugar beet transmitted the virescence agent to periwinkle.

Thus, all leafhoppers that fed on the periwinkle plants that developed virescence also fed on the sugar beet plants, but some of the sugar beet plants did not develop curly top. Although the curly top virus was present in some of the original periwinkle plants with symptoms of virescence, it did not seem to be the agent that caused virescence.

To test further whether curly top virus caused virescence in periwinkles, we exposed groups of *C. tenellus* known to carry curly top virus to 30 healthy periwinkle plants and allowed the leafhoppers to feed until they died. After several months, none of the 30 plants had developed virescence or any other detectable symptoms of any disease.

Then we exposed *C. tenellus* known to be free of curly top virus to 3 of the 30 plants for one week and transferred them to healthy sugar beets. Within two weeks, all the sugar beets developed curly top, which indicated that the leafhoppers had acquired curly top virus from the apparently healthy periwinkle plants and also that curly top virus invades periwinkle but does not cause symptoms of virescence or of any other disease.

On the basis of these tests, we concluded that the agent causing virescence that is harbored by *C. tenellus* in the field and is transmissable by laboratory-reared *C. tenellus* is neither *S. citri* nor curly top virus.

We next cut thin sections of the stems of healthy and virescence-affected periwinkle plants, stained them, and examined them with an electron microscope. The phloem (food conducting tissue) of those plants with virescence contained many mycoplasma-like bodies; such bodies were not present in phloem from healthy periwinkles. Also, we were able to transmit virescence to healthy periwinkle plants by grafting pieces of stem from affected periwinkles.

We fed laboratory-reared *C. tenellus* on a dooryard periwinkle plant from Riverside that exhibited virescence, and then fed them on healthy periwinkles in the laboratory. Virescence developed on the laboratory plants. Again in 1975 and 1976, *C. tenellus* collected from the field harbored and transmitted virescence to healthy periwinkle plants.

Other species of leafhoppers collected in the field have not transmitted virescence to healthy periwinkle. Even when one extensively collected species, *S. nitridus*, was fed on virescence-infected periwinkle in the laboratory, the insects failed to transmit virescence when subsequently fed on healthy periwinkle.

Causal agent

The virescence agent, apparently a phloem-inhabiting mycoplasma-like organism, is naturally harbored by *C. tenellus* collected from several locations in California. Such mycoplasma-like organisms cause disease in many agriculturally important plants, and we are endeavoring to determine whether this agent causes disease in other plants—either agriculturally important plants or wild species that may act as natural reservoirs.

George N. Oldfield is Research Entomologist, George H. Kaloostian is Research Leader, and Harold D. Pierce is Agricultural Research Technician, USDA, ARS, Boyden Entomology Laboratory, Riverside; Andrew L. Granett is Post-graduate Research Plant Pathologist, Statewide Air Pollution Research Center, and Edmond C. Calavan is Professor and Plant Pathologist, Department of Plant Pathology, University of California, Riverside.