

**Identification of Citrange Stunt as the Mechanically Transmissible Virus from Meyer Lemons Doubly Infected with Citrange Stunt and Tatter Leaf Viruses**

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Wallace & Drake (4) described as "tatter leaf" a virus disease produced on *Citrus excelsa* Wester graft-inoculated from plants of Meyer lemon (*C. limon* × *C. sinensis*?). Later, Wallace & Drake (5) reported that citrange (*Poncirus trifoliata* × *C. sinensis*) and citremon (*P. trifoliata* × *C. limon*) developed strong leaf effects and stem pitting when inoculated from Meyer lemons that carried tatter leaf virus (TLV). It was concluded that the symptoms on *C. excelsa* and the trifoliolate hybrids were caused by the same virus.

Yarwood (7) reported mechanical transmission of a virus from Meyer lemon which produced local lesions and systemic infection in cowpea (*Vigna sinensis* [Torner] Savi), bean (*Phaseolus vulgaris* L.), cucumber (*Cucumis sativus* L.), and some other plants. Semancik & Weathers (3) mechanically transmitted virus from a tatter leaf-infected *C. excelsa* to Troyer citrange which they concluded was TLV. With the same inoculum they produced diffuse necrotic lesions on cowpea, and by subinoculations from infected cowpea they obtained infection in 19 plant species, including lime (*C. aurantifolia* [Christm.] Swingle), which became infected but did not display symptoms. In partially purified preparations from cowpea mechanically inoculated from infected *C. excelsa*, Semancik & Weathers (3) observed filamentous particles averaging about 650 m $\mu$  in length with a uniform width of 19 m $\mu$ . They emphasized that transmission from infected herbaceous hosts to suitable citrus indicator hosts and production of symptoms of tatter leaf were needed to confirm that this virus was TLV.

Fulton (1) compared cultures of virus studied by Yarwood (7) and Semancik & Weathers (3). He observed no difference between these cultures, both of which were mechanically inoculated from infected cowpea to rough lemon (*C. jambhiri* Lush.). These rough lemon plants developed no symptoms, but graft inoculations from them to citrange and citremon resulted in symptoms typical of those described originally by Wallace & Drake (5), who at first assumed that they were caused by TLV.

Wallace & Drake (6) presented evidence that two viruses are present in Meyer lemon plants which are carriers of tatter leaf virus. They demonstrated that plants of *C. excelsa* which recovered from tatter leaf symptoms after graft inoculation from Meyer lemon

no longer contained virus which caused tatter leaf symptoms on *C. excelsa*. The fact that the recovered *C. excelsa* plants again developed tatter leaf symptoms when reinoculated from Meyer lemon was additional evidence that recovery from symptoms was associated with loss of tatter leaf virus. However, graft inoculations from these recovered *C. excelsa* plants to Troyer citrange and citremon resulted in the previously described symptoms on these hosts. Consequently, the name "citrange stunt" was given to the disease effects on citrange and citremon by Wallace & Drake (6), who stated that Semancik & Weathers (3) apparently mechanically transmitted citrange stunt virus (CSV), but not the virus in Meyer lemon which causes tatter leaf symptoms on *C. excelsa*.

The paper by Wallace & Drake (6) was presented at a conference on citrus virus diseases in 1966, and was prepared before the report of Fulton (1) appeared in print. Fulton (1) confirmed that the virus which is transmissible from Meyer lemon to cowpea causes the disease effects on citrange and citremon, and he assumed that these were caused by TLV. With the information available to him that assumption was reasonable, but it is now known that to identify the virus as TLV, back-inoculations from the infected citrange and citremon should have been made to *C. excelsa* with production of symptoms of tatter leaf on that host.

It should also be mentioned that the studies by Garnsey & Jones (2) on distribution of virus in affected leaves of Rusk citrange dealt with CSV and not TLV. Their paper, like that of Fulton (1), was submitted for publication before identification of CSV had been made.

Evidence of the presence of two distinct viruses in Meyer lemon was obtained by Wallace & Drake (6) entirely through tissue graft inoculations. This paper describes results of additional transmission experiments which provide proof that CSV, not TLV, was mechanically transmitted and partially purified by the investigators mentioned above.

Meyer lemon 1226, one of the original sources of TLV, and Troyer citrange seedling 1768 were used as sources of inocula. Inoculation by grafting from Meyer lemon 1226 had given symptoms of tatter leaf on *C. excelsa* and typical symptoms of citrange stunt on Troyer citrange. Troyer seedling 1768, showing symptoms of citrange stunt, had been infected with virus from Meyer lemon 1226 after serial passage through (i) *C. excelsa* which developed tatter leaf and recovered; (ii) Troyer citrange which developed the symptoms now identified as citrange stunt; and (iii) Mexican lime which remained symptomless. Back-inoculations by tissue grafts from Troyer 1768 had given no symptoms on *C. excelsa*. This test was repeated by grafting small stem pieces from Troyer 1768 to seedlings of *C. excelsa* and Troyer citrange. Within 1 month, leaves of inoculated Troyer plants developed typical citrange stunt symptoms. The inoculated *C. excelsa* seedlings remained free of symptoms. This again demonstrated that Troyer 1768 contained no

virus capable of causing tatter leaf on *C. excelsa*, but was infected with CSV.

Primary leaves of cowpea var. Early Ramshorn were sap-inoculated from Meyer lemon 1226 and Troyer citrange 1768. Inocula consisted of macerates of young leaves and flower petals of Meyer lemon and mature leaves of Troyer citrange in potassium phosphate buffer (0.02 M at pH 7.5). Plants inoculated from both sources developed reddish, local necrotic lesions 3-4 days after inoculation. Systemic mottle and reddish stem necrosis appeared later. Since Meyer lemon 1226 carried both TLV and CSV, it was to be expected that cowpea plants would react to inoculation from it. The fact that an identical reaction was obtained on cowpea sap-inoculated from Troyer citrange 1768 which was not infected with TLV establishes that it is CSV that has been mechanically transmitted from citrus to herbaceous hosts and back to citrus. Currently, the only known reliable indicator host of TLV is *C. excelsa*. Thus, to demonstrate mechanical transmission of TLV will require the production of tatter leaf symptoms on this host by sap-inoculation directly from known infected citrus, from sap-inoculated herbaceous plants, or by graft-inoculation from

citrus which may have been infected by sap-inoculation from a TLV source.

## LITERATURE CITED

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