Natural Occurrence of Tomato Ringspot Virus in *Passiflora edulis* from Peru

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ABSTRACT

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An isometric virus isolated from *Passiflora edulis* in Peru had typical properties of a nepovirus. It infected a wide range of dicotyledonous plants, inducing mainly chlorotic and/or necrotic spots and rings. In *Chenopodium* species, it caused apical necrosis. It was not transmitted by aphids. Its coat protein had a molecular weight of about 56,000. In the serological agar gel double-diffusion test, it could not be distinguished from the tobacco strain of tomato ringspot virus. Unlike the type strain of this virus, however, it did not infect tomato systemically.

From passion fruit plants (*Passiflora edulis* Sims; Spanish: maracuyá) grown in the coastal area of northern Peru that showed mosaic and ringspot symptoms, two viruses were isolated: a tobamovirus named maracuja mosaic virus (MrMV; C. E. Fribourg, R. Koenig, and D. E. Lesemann, *unpublished*) and an isometric virus identified as tomato ringspot virus (TomRV).

MATERIALS AND METHODS

The virus isolated from passion fruit was separated from MrMV by five successive single-lesion transfers to *Chenopodium quinoa* Willd. and was then maintained on *Nicotiana benthamiana* Domin. Transmission experiments were done in an insectproof greenhouse. Aphid transmission was attempted with *Myzus persicae* (Sulz.) reared on *Brassica pekinensis* (Lour.) Rupr. The aphids were starved for 1 hr, then allowed to feed on infected *N. benthamiana* for 30–60 sec before being transferred to healthy *N. benthamiana* seedlings. Seed transmissibility of the virus was checked by rubbing *C. quinoa* plants with sap from seedlings that had been grown from seed produced by infected *N. benthamiana* or *Vigna unguiculata* (L.) Walp.

The virus was partially purified by the method of Stace-Smith (15). The protein molecular weight was determined on 10% polyacrylamide gels according to the methods of Laemmli and Favre (13). Serological agar gel double-diffusion tests were done with 0.85% Difco Noble agar containing 0.85% sodium chloride, 0.25% sodium azide, and 0.01 M Tris-HCl buffer, pH 8.0.

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RESULTS

Passion fruit virus isolate MV had a wide host range, infecting species belonging to the families Amaranthaceae, Balsaminaceae, Chenopodiaceae, Compositae, Cucurbitaceae, Labiatae, Leguminosae, Pedaliaceae, Passifloraceae, Portulacaceae, and Solanaceae, inducing mainly chlorotic and/or necrotic spots or rings. The following hosts were systemically infected after mechanical inoculation: *Amaranthus edulis* L., *Gomphrena globosa* L., *Impatiens balsamina* L., *Chenopodium amaranticolor* Coste & Reyn., *C. murale* L., *C. quinoa*, *Helianthus annuus* L., *Helichrysum bracteatum* Andr., *Zinnia elegans* Jacq., *Cucumis sativus* L., *Cyclanthera pedata* Schrad., *Salvia splendens* Ker-Gawl, *Clitoria ternatea* L., *Phaseolus aborigineus* Burkart (PI 266-910), *P. acutifolius* Gray (PI 310801), *P. vulgaris* L. 'Monroe,' *V. unguiculata* subsp. *unguiculata* 'Black,' *V. unguiculata* subsp. *cylindrica* 'Catjang,' *Passiflora edulis* Sims., *Sesamum indicum* L., *Montia perfoliata* (Willd.) Howell, *Datura stramonium* L., *Nicandra physaloides* Gaertn., *Nicotiana benthamiana*, *N. debneyi* Domin., *N. tabacum* L. 'Samsun,' and *Physalis floridana* Rybd. The virus induced characteristic symptoms of necrotic local lesions followed by top necrosis in *Chenopodium amaranticolor* and *C. quinoa*; systemic chlorotic spots and rings in *Cucumis sativus*; local and systemic necrotic spots in *Phaseolus vulgaris* 'Monroe'; necrotic local spots and rings followed by systemic line patterns in *Nicotiana tabacum* 'Samsun'; and local necrotic spots, partial stem necrosis, and chlorotic or necrotic spots in trifoliate leaves of *V. unguiculata*. Tomato cultivars Marglobe, San Marzano, Stone, VF-145, and UC-82 developed local necrotic concentric rings or chlorotic spots, but the virus did not become systemic. Mechanically inoculated passion fruit plants showed systemic necrotic spots and chlorotic line patterns during the spring but were symptomless during the summer.

Local infection without systemic movement of the virus was observed in *Cucurbita maxima* Duch., *C. pepo* L., *Lagenaria siceraria* (Molina) Standl., *Luffa acutangula* Roxb., *P. vulgaris* L. 'Pinto,' *Datura metel* L., *Lycopersicon pimpinellifolium* (Jusl.) Mill., *N. glutinosa* L., and *Solanum tuberosum* subsp. *tuberosum* × subsp. *andigena* 'Mariva,' 'Tomasa Condemayta,' and 'Yungay.'

The infectivity of the virus in sap from *N. benthamiana* was lost by dilution at 10^{-5} but not 10^{-4}, by heating at 60 but not 55 C, and by storage at about 22 C for 9 but not 7 days. The virus was not transmitted by seed of *N. benthamiana* or *V. unguiculata* or by *M. persicae* when acquisition feeding of 30–60 sec was allowed.

In purified virus preparations, isometric particles with diameters between 25 and 30 nm were seen after staining with uranyl acetate. The preparations contained a large proportion of particles that were penetrated by the stain.

In SDS polyacrylamide gel electrophoresis, the virus yielded three closely spaced protein bands for which molecular weights between 55,000 and 57,000 were calculated.

Host reactions, particle morphology, and the unusually high protein molecular weight suggested that the virus was a nepovirus. Agar gel double-diffusion

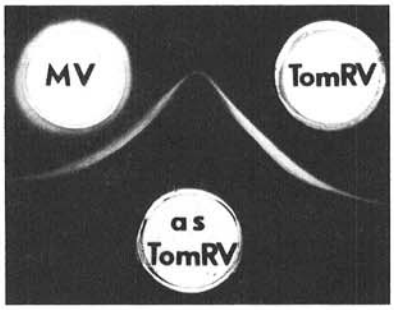

Fig. 1. Reaction of the passion fruit virus isolate MV with an antiserum to the tobacco strain of tomato ringspot virus (TomRV) and lack of spur formation with this isolate of TomRV.

tests were done with antisera to arabis mosaic, grapevine fanleaf, potato virus U, tobacco ringspot, tomato black ring viruses, and the tobacco strain (16) of TomRV. Only with the antiserum to TomRV, a strong precipitin line formed that was confluent with that given by the homologous virus (Fig. 1). The lines formed by the two isolates with an antiserum to the passion fruit isolate MV also fused without spur formation.

DISCUSSION

The isometric virus isolated from passion fruit has proven to be TomRV. Its host range and symptomatology in the diagnostic species *C. amaranticolor, C. sativus, P. vulgaris, N. tabacum*, and *V. unguiculata* are similar to those of other TomRV isolates (6,7,10,16). This virus is known to cause diseases in a number of woody plants, but passion fruit had not been reported previously to become infected.

TomRV is widespread in North America (16) and has also been reported from other parts of the world, e.g., Australia (2), New Zealand (5), and the Far East (8,10,17). In Europe, it has been found occasionally, e.g., in Pelargonium (1,9,14) and raspberry (11), but the infection has usually been traced back to imports from abroad (9). The nematode vector of the virus, *Xiphinema americanum* Cobb, is not native to Europe.

TomRV has not been reported previously from Peru and has only been reported once from the South American continent (3). It may, however, well be native to Peru. Its vector, *X. americanum*, is widespread in this country (12), where it may transmit the calico strain of tobacco ringspot virus (4; C. E. Fribourg and P. Jatala, *unpublished*). Alternatively, it is also possible that it has been introduced in fruits, trees, shrubs, or grapevines to the cool, hilly areas of northern Peru, and once established, it has spread by a combination of seed and vector transmission to the coastal region where passion fruit is grown.

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