Manual Transmission of Dasheen Mosaic Virus from *Richardia* to Nonaraceous Hosts

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ABSTRACT

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Dasheen mosaic virus was isolated from diseased calla lily plants (*Richardia africana*) growing in southern Italy. *Chenopodium amaranticolor, C. quinoa, C. ambrosioides, Nicotiana benthamiana,* and *Saponaria vaccaria* were infected when inoculated with triturated epidermal strips from *R. africana*.

Additional key words: Araceae, *Zantedeschia* sp.

Plants of *Richardia africana* (=*Zantedeschia aethiopica*) with foliar distortion and discoloration (mosaic, mottle, and yellowing) (Fig. 1) were observed in gardens near Bari in southern Italy. Symptoms were most conspicuous in the youngest leaves during winter and spring. A marked asymmetry of some leaves was also observed. This paper reports on the transmission of dasheen mosaic virus (DMV) from these plants and on several new nonaraceous suscepts of this virus.

MATERIALS AND METHODS

Test plants dusted with Celite were inoculated manually with infected leaf tissue from *R. africana*. Either leaf pieces or epidermal strips were triturated with a mortar and pestle using 0.2 M phosphate buffer (pH 7.0) containing 1% sodium ascorbate as the diluent. After inoculation, the plants were rinsed with tap water and placed in a shaded greenhouse at 18–24 C for observation.

Epidermal strips from lower leaf surfaces of naturally infected *R. africana* leaves were stained with calcomine orange and Luxol brilliant green and examined with a light microscope for viral inclusions (2). Leaf extracts were negatively stained in phosphotungstate (pH 6.8) and examined for virus particles with a Philips model 201 C electron microscope.

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The following serological techniques were used: immunodiffusion and immunosorbent and decoration electron microscopy (10). The immunodiffusion medium consisted of 0.8% Noble agar, 0.2% sodium dodecyl sulfate, 0.1% NaN_3, and 0.7% NaCl (12). The antiserum used was that described by Abo El-Nil et al (1).

RESULTS

None of the tests plants developed symptoms when triturated leaf pieces of *R. africana* were used as the inoculum, but when triturated epidermal leaf strips from this host were used, local chlorotic lesions developed on leaves of *Chenopodium amaranticolor, C. quinoa* (Fig. 2A), *C. ambrosioides, Nicotiana benthamiana*, and *Saponaria vaccaria* 'Pink Beauty' 10–15 days after inoculation. In some instances, *C. quinoa* and *N. benthamiana* plants also developed systemic symptoms consisting of yellowing of secondary veins and pinpoint chlorotic spots (Fig. 2B). In *S. vaccaria*, poorly defined chlorotic local lesions were noted, but attempts to recover the virus from these hosts by back-inoculations to *Philodendron selloum* seedlings were unsuccessful.

No symptoms developed in the following plants, which were similarly inoculated: *Beta vulgaris, Cucumis sativus* 'Marketer,' *Cucurbita pepo, Datura stramonium, Gomphrena globosa,*

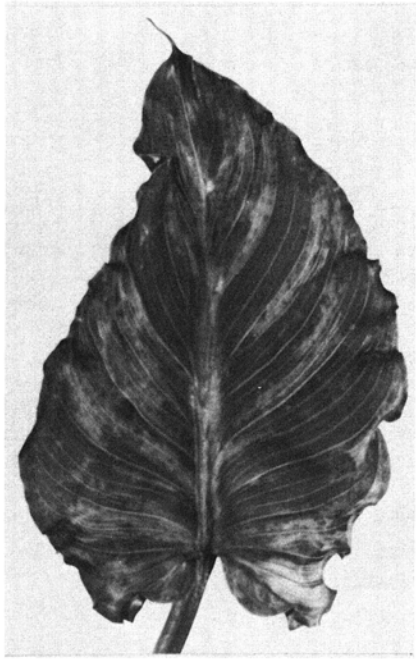

Fig. 1. Naturally infected leaf of *Richardia africana* showing foliar mosaic and distortion symptoms.

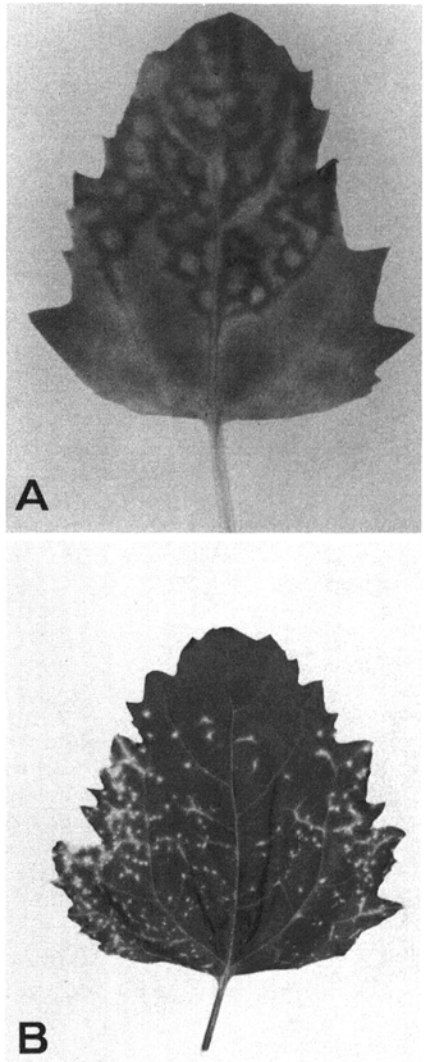

Fig. 2. (A) Local and **(B)** systemic symptoms of *Chenopodium quinoa* leaves 3 and 5 wk, respectively, after manual inoculation with dasheen mosaic virus.

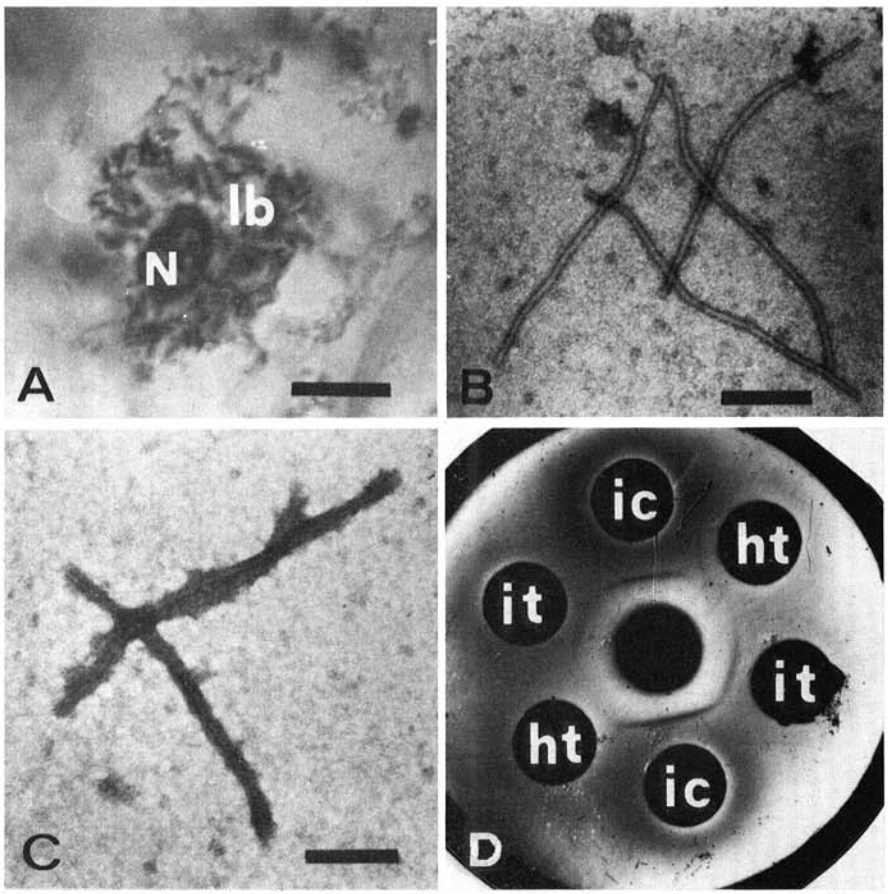

Fig. 3. (A) Cytoplasmic inclusion (Ib) in an epidermal cell of *Richardia africana* naturally infected with dasheen mosaic virus (DMV). N = nucleus. Scale bar = 10 μm. **(B)** Flexuous-rod virus particles from negatively stained extracts of manually inoculated *Chenopodium quinoa* leaves. Scale bar = 200 nm. **(C)** Flexuous-rod virus particles from *C. quinoa* leaves decorated by DMV antiserum. Scale bar = 200 nm. **(D)** Serological reactions in immunodiffusion tests. Center well contains DMV antiserum and peripheral wells contain antigens treated with one volume of 3% sodium dodecyl sulfate; it = homologous DMV-infected taro (*Colocasia esculenta*) leaf extract, ic = heterologous infected *R. africana* leaf extract, and ht = healthy taro.

N. clevelandii, N. glutinosa, N. rustica, N. tabacum 'White Burley' and 'Samsun,' *Phaseolus aureus*, and *P. vulgaris* 'La Victoire.'

Cylindrical inclusions characteristic of DMV and other potyviruses (2) were observed in the cytoplasm of *R. africana* epidermal cells (Fig 3A).

Flexuous-rod virus particles were observed in negatively stained leaf extracts of *R. africana, C. amaranticolor, C. quinoa, C. ambrosioides, N. benthamiana,* and *S. vaccaria* leaves with symptoms (Fig. 3B). Also, in decoration experiments, DMV antibody was specifically adsorbed by virus particles extracted from leaves of *R. africana* and *C. quinoa* (Fig. 3C).

In unilateral immunodiffusion tests, R. africana leaf extracts reacted identically with DMV antiserum (Fig. 3D), as did leaf extracts of *C. quinoa.*

DISCUSSION

Particle morphology, cytoplasmic inclusions of the virus infecting *R. africana*, and serological studies all provide evidence that the virus is DMV (14). Although this appears to be the first report of DMV in Italy, this virus has been found elsewhere in Europe (5,6,11,13). Also, a similar virus from *R. africana* has been described from the Soviet Union (8), although its relationship to DMV is unknown (14). Lovisolo and Conti (9) reported natural infection of the aroid *Arum italicum* by cucumber mosaic virus in Italy, but no evidence for this virus was obtained in our tests.

With the possible exception of *Tetragonia expansa* (14; A. A. Brunt, *personal communication*), DMV has been considered to be restricted to the Araceae. As shown in this study, using triturated epidermal strips of *R. africana* as inoculum, DMV can also infect plants in other families, including the Chenopodiaceae, Solanaceae, and Caryophyllaceae. It is not clear why epidermal tissues are better inoculum sources than intact leaves, but possible inhibitory substances in subepidermal tissue may be responsible. *S. vaccaria, N. benthamiana,* and *Chenopodium* spp., which are susceptible to many different viruses (3,4,7), may be useful as indicator plants for DMV. It is unlikely, however, that they will be epidemiologically significant in the spread of this virus because, in most instances, only localized infections have resulted. The low virus titers supported by these hosts may explain the failure to recover DMV.

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