CRISTACORTIS, IMPIETRATURA, and RELATED DISEASES

Studies on Impietratura Disease

G. Cartia and A. Càtara

Impietratura of citrus has been studied extensively, but many questions remain unanswered (*2*). This paper reports results of studies on natural spread in the field, histological changes in fruits and twigs of inoculated citrus, and the suspected relation of impietratura to rumple disease of lemon (*8*).

MATERIALS AND METHODS

Investigations on natural spread of the disease were started in 1964 in a Sanguinello orange orchard where the disease had been noticed many years before. The orchard consisted of 360 20-year-old trees, spaced 4 × 4 m. Trees and ripened fruits were inspected yearly. Fruits suspected of being affected were peeled to show internal symptoms.

Histological studies were made on the following citrus plants: *Citrus volkameriana* and Ovale orange, inoculated and showing gum in the albedo; sour orange, citron, and kumquat, inoculated but symptomless; naturally-infected Tarocco orange showing dark-green areas without gum in the albedo, ascribed to impietratura infection (*7*); Sanguinello budlings inoculated six years before; and Femminello lemon exhibiting typical symptoms of rumple. Fruits, pedicels, twigs, leaf blades, and petioles were sectioned and stained according to the thionin-orange G procedure (*3, 6*), which stains secreted, gumlike material in the tissue a deep blue, necrotic tissue a bright blue, and normal tissues a yellow-orange.

Transmission tests with rumple were tried on trees of two nucellar clones of lemons grafted on *Citrus volkameriana*. Bark tissues of trees with rumpled lemons were grafted on branches and twigs of 19 trees, leaving four trees as check plants. Fruits were examined periodically for symptoms until they reached full size; they were then picked, peeled, and examined internally.

RESULTS

Field observations. During the first three years (1964–1966) the number of affected trees increased, suggesting natural spread, as reported earlier by Scaramuzzi *et al.* (*5*). In the last six years (1967–1972), however, there was no increase in the number of trees showing impietratura, suggesting no spread.

Histological studies. Affected fruits of susceptible citrus, such as *C. volkameriana* and Ovale orange, showed anatomical change before any external symptom was recognizable. The change was necrosis of vascular bundles of peel, which stained deep blue, much like symptoms observed by Safran (*4*) on Shamouti orange and grapefruit. The more the fruit was affected, the more severe the necrosis that appeared in cells of the albedo. In young fruits in advanced stages of the disease, irregular cavities could be seen, lined by parenchymatous cells surrounding small islands of normal tissue. Later, the cells

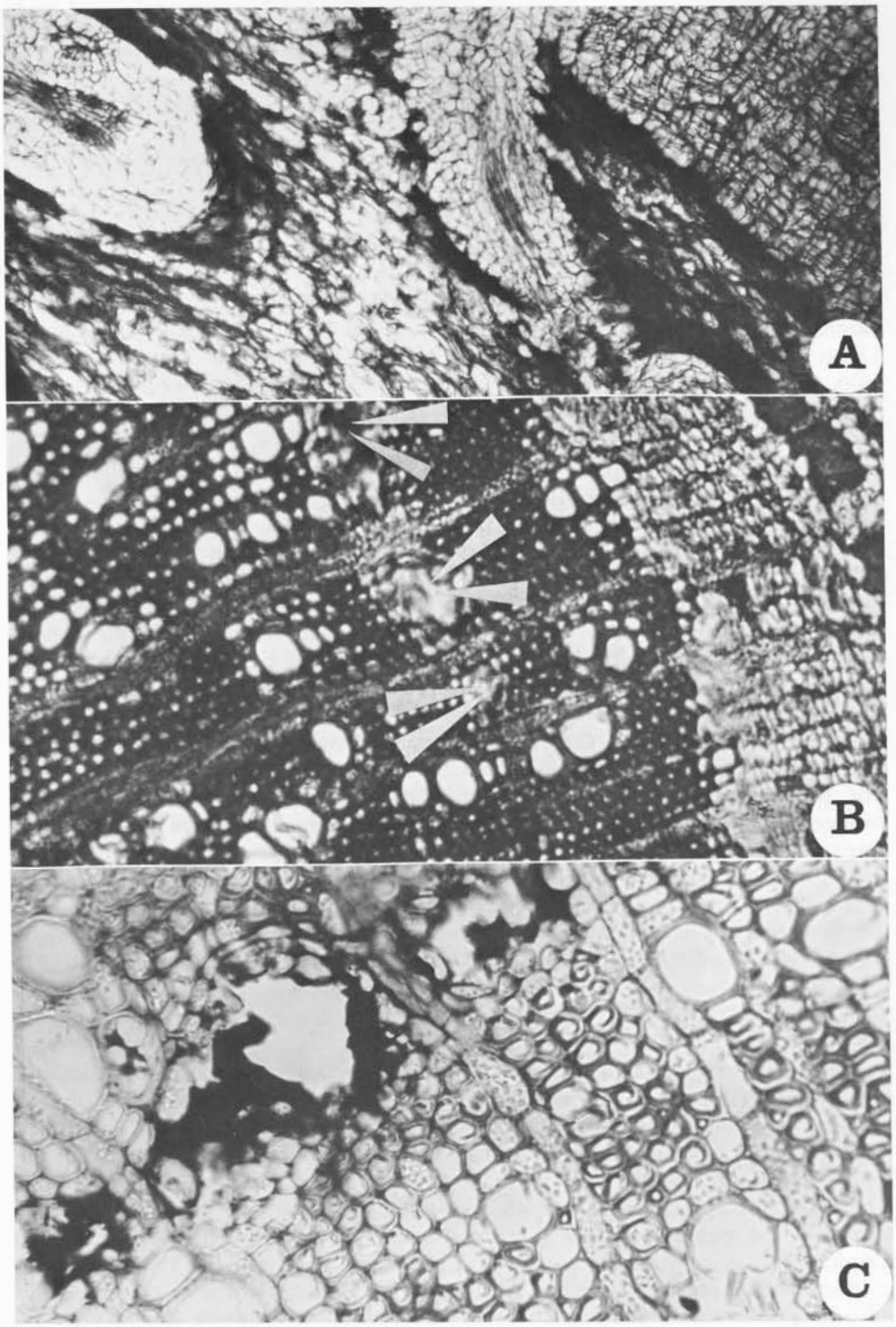

Fig. 1. Histological changes associated with impietratura in *Citrus volkameriana*. *A*, radial section through the albedo, showing necrotic cells and gumlike material, which stains blue. *B*, cross-section through a young twig: arrows indicate vessels filled with gumlike material. *C*, cross-section of a pedicel showing gumlike material in the vessels.

lining the cavities secreted a gumlike substance that filled the cavities (fig. 1A).

In affected pedicels, histological changes were observed only in xylem. Vascular rays and the cambium remained normal. Some vessels of xylem stained deep blue and were easily recognizable (fig. 1C).

Similar symptoms were detected in one to three-year-old twigs of affected trees. The gumlike material was observed mainly in the spring or summer rings of xylem, and more than one ring was affected (fig. 1B).

No symptoms were observed in the leaf blades and petioles of affected *Citrus volkameriana*.

In trees showing no external symptoms, such as citron, sour orange, and kumquat, only a few vessels in the twigs were stained. Changes in the fruits consisted only of slight necrosis of vascular bundles.

Tarocco orange fruits with dark-green areas showed deep blue-stained vascular bundles and some necrotic, compact cells below the light-colored, depressed areas.

Sanguinello twigs showed only a mild reaction in the xylem.

Necrosis of the vascular bundles or other internal modifications were not evident in rumple-affected lemon fruits. Epidermal layers were unaffected even though rinds were heavily corrugated. Pedicels and twigs of rumple-affected lemons did not show any of the described symptoms in the xylem.

Transmission tests. Among 235 lemon fruits picked from trees inoculated with rumple, 10 fruits showed typical symptims of rumple and six showed atypical symptoms. Eight fruits of *Citrus volkameriana* borne on suckers of the rootstock of two inoculated plants showed no symptoms of either rumple or impietratura. Forty fruits from check plants showed no rumple.

DISCUSSION AND CONCLUSIONS

Results reported herein agree with other observations on impietratura, indicating that no natural transmission takes place from diseased to healthy plants (*2*). Variability of symptoms of the disease could be related to differences in pathogen, susceptibility of the host, and environment (*1*), and could explain suspected natural spread reported by us and others (*2, 5*).

Histological similarities of impietratura in twigs of susceptible and symptomless citrus, that appear different from those reported for other citrus virus diseases, might be a helpful tool in indexing for impietratura.

Results do not confirm any relation between rumple of lemon and impietratura. This conclusion is based on histological differences and the failure to induce rumple or impietratura symptoms in transmission tests.

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