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Stunting, Budunion Crease, and Inverse Pitting of Grapefruit on Sour Orange Rootstock

L. W. Timmer and S. M. Garnsey

ABSTRACT. A budunion disorder was observed on severely stunted grapefruit trees on sour orange rootstock in five Texas orchards. Affected trees had a severe crease at the budunion which was sometimes associated with pitting and gum impregnation of the wood on the sour orange rootstock. Inverse pitting on the grapefruit scion was observed on several trees. Some initial serological and cytological tests were positive for citrus tristeza virus, but they could not be confirmed by indexing or further testing. The possibility that the disorder was due to the effect of xyloporosis on off-type sour orange rootstocks was not eliminated. Similar symptoms occurred on some budwood propagations from affected trees on sour orange rootstock, but many propagations appeared healthy. Only 1 of 10 healthy grapefruit on sour orange rootstock developed a budunion crease following inoculation from a diseased field tree.

Index words. enzyme-linked immunosorbent assay, inclusion bodies.

Scattered, severely stunted grapefruit trees were observed in several citrus orchards in the Lower Rio Grande Valley in Texas in 1972. These trees, reportedly propagated on sour orange rootstock, showed obvious budunion problems not readily attributable to any known citrus pathogen. However, symptoms were reminiscent of those caused by citrus tristeza virus (CTV) which has never been a problem in Texas citrus orchards. Thus, the disorder represented a serious potential threat to the Texas citrus industry. In this paper, we report the symptoms and occurrence of the disorder and attempts to determine its cause.

FIELD SYMPTOMS

Trees in several orchards where growers reported stunting of young grapefruit trees on sour rootstock (fig. 1A, B) were examined for budunion and wood abnormalities by removing bark from the budunion and from various sized branches. The most common symptom observed was an overgrowth of the scion with a distinct crease and pitting at the budunion (fig. 1B, inset; fig. 2A, B).

When a strong crease occurred at the budunion, there was usually only mild pitting on the sour orange rootstock (fig. 2A, B) and the grapefruit scion was unaffected. On a few trees, gum impregnation was observed in the rootstock bark and some gum was associated with the pits. The budunion crease was the most common symptom on stunted trees and was observed in all five orchards (table 1). Only a few scattered trees were affected in each orchard.

In a few less severely stunted trees, we observed inverse pitting on the grapefruit scion with no apparent problem at the budunion (fig. 2D). Pegs were observed even on the smaller branches and bark was severely pitted (fig. 2E). This disorder was observed on only four trees in two orchards (table 1). One tree in orchard C had both the budunion crease and the inverse pitting on grapefruit.

ETIOLOGY

Since the symptoms at the budunion were somewhat similar to those produced in trees on sour orange rootstock by citrus tristeza virus (CTV), we indexed for CTV using several methods. In initial tests, serological techniques (1, 4) were used to test for CTV in twig.
other graft-transmissible agents

Fig. 1. Field symptoms of the budunion crease problem of grapefruit on sour orange. A, B) Stunted grapefruit tree on sour orange rootstock with a budunion crease; healthy tree in the background in A; (inset) close-up of budunion of the tree in B.

Bark from stunted trees. Five of 28 affected trees assayed by sodium dodecyl sulfate (SDS)-immunodiffusion and 8 of 34 trees tested by enzyme-linked immunosorbent assay (ELISA) gave weak positive reactions. Three other sources known to be infected with CTV gave strong positive reactions and eight known healthy sources did not react in these tests. Root bark from six affected trees gave moderate positive reactions in ELISA tests. Staining for CTV inclusion
bodies (3) in leaf petioles from affected trees yielded questionable positives in 15 of 34 cases. In extensive subsequent tests, however, we were unable to confirm CTV infection of these sources by SDS-immunodiffusion, ELISA, or by staining for CTV inclusion bodies.

In indexing tests, twig bark from 25 affected trees was graft inoculated into 1-3 Mexican lime seedlings per source. In addition, three Mexican lime seedlings were side-grafted with roots from each of three affected trees in the field. None of the inoculated Mexican lime seedlings developed vein clearing, stem pitting, or other CTV symptoms.

Bark from two sources affected with budunion crease in orchard C was grafted into three seedlings each of sour orange, Troyer citrange, Pineapple sweet orange, *Citrus excelsa*, and Hudson grapefruit. After 1.5 years in the greenhouse, none of these plants developed symptoms of any known virus disease or other viruslike symptoms and all grew normally.

We also considered the possibility that the symptoms might be due to the effect of xyloporosis on misidentified, mislabeled, or off-
type rootstock seedlings. Xyloporosis is commonly carried by old-line bud sources in Texas (12), and trees in orchards A, B, C, and D had been propagated from old-line sources. Trees in at least three orchards (A, B, C) were obtained from the same nursery and the source of trees for orchard D was unknown. The one tree in orchard E was presumably of nucellar origin, but this could not be confirmed.

Many affected trees were cut off below the budunion to force rootstock sprouts for identification. Most of these trees were in such poor condition that they failed to sprout. Of the five that sprouted, two had typical sour orange foliage and the other three appeared to be off-types. One of these had very narrow, strap-shaped leaves, one produced some trifoliate leaves, and the third had lemonlike leaves with narrow wings on the petioles. Thus, it appeared that the rootstock had been grown from a sour orange seed source, but that at least some off-type seedlings had been budded.

**PROPAGATION AND TRANSMISSION**

Propagations of buds from 5 of the 10 trees affected by budunion crease produced trees with abnormalities (table 2). Only 3 of the 10 produced symptoms which were typical of girdling or budunion problems. The two others had abnormally thick leaves similar to those on tetraploid grapefruit seedlings. The rest of the budwood propagations remained healthy for 2-3 years in the greenhouse. Six trees transplanted to the field have remained healthy for 5-6 years.

Attempts to transmit the disorder to healthy, young grapefruit on sour orange rootstock, produced only one plant with symptoms (table 2). In this case, the bud produced a normal-appearing shoot about 25 cm long. As the second flush of growth occurred, the mature leaves developed veinal chlorosis and the new shoot lost leaves and died back. This tree had a crease at the budunion (fig. 1C).

**DISCUSSION**

The disorder reported here could not be definitely associated with a known citrus virus. Since occasional samples tested positive for CTV by serological and cyto logical methods, the disease could be due to CTV which we failed to graft transmit to indicator plants. Erratic distribution of CTV in grapefruit trees has been observed previously (4; Cohen and Garnsey, unpublished). We also cannot eliminate the possibility that it is an effect of xyloporosis virus on an off-type source orange rootstock seedling. The single graft transmission observed indicated that the disorder may be due to an in-

### TABLE 1

<table>
<thead>
<tr>
<th>Orchard</th>
<th>Age (year)</th>
<th>No. trees affected/no. examined</th>
<th>Symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>6-7</td>
<td>8/8</td>
<td>Budunion crease</td>
</tr>
<tr>
<td>B</td>
<td>2-3</td>
<td>6/6</td>
<td>Budunion crease</td>
</tr>
<tr>
<td>C</td>
<td>3-4</td>
<td>11/13</td>
<td>Budunion crease</td>
</tr>
<tr>
<td>D</td>
<td>3-4</td>
<td>8/9</td>
<td>Inverse pitting</td>
</tr>
<tr>
<td>E</td>
<td>4-5</td>
<td>1/9</td>
<td>Budunion crease</td>
</tr>
</tbody>
</table>

SYMPTOMS OBSERVED ON STUNTED GRAPEFRUIT TREES ON SOUR ORANGE ROOTSTOCK IN SEVERAL TEXAS ORCHARDS

Orchard, Age (year), No. trees affected/no. examined, Symptoms.
TABLE 2
ATTEMPTS TO PROPAGATE AND GRAFT-TRANSMIT THE BUDUNION CREESE OF GRAPEFRUIT ON SOUR ORANGE ROOTSTOCK

<table>
<thead>
<tr>
<th>Source Orchard Tree</th>
<th>No. positive/ no. tested</th>
<th>Symptom</th>
</tr>
</thead>
<tbody>
<tr>
<td>Propagations A</td>
<td>3/3</td>
<td>Apical necrosis, death</td>
</tr>
<tr>
<td></td>
<td>2/3</td>
<td>Epinasty, bloom, veinal chlorosis</td>
</tr>
<tr>
<td>B</td>
<td>0/3</td>
<td>None</td>
</tr>
<tr>
<td>C</td>
<td>0/3</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>0/3</td>
<td>Thickened leaves</td>
</tr>
<tr>
<td></td>
<td>0/3</td>
<td>Thickened leaves</td>
</tr>
<tr>
<td></td>
<td>0/3</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>0/3</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>0/3</td>
<td>Epinasty, bloom, budunion crease</td>
</tr>
<tr>
<td>Transmission* C</td>
<td>0/5</td>
<td>None</td>
</tr>
<tr>
<td></td>
<td>1/5</td>
<td>Veinal chlorosis, leaf drop, budunion crease</td>
</tr>
</tbody>
</table>

*Healthy grapefruit on sour orange rootstock graft inoculated with tissue from affected trees.

fectious agent rather than to a genetic abnormality. Genetic abnormalities are usually consistently propagated, whereas infectious agents are not always because of irregular distribution of the pathogen (4, 16).

A number of other budunion abnormalities have been described on various rootstock-scion combinations. Some disorders such as those involving various combinations with kumquat and Calamondin (10, 11) and Eureka lemon budded on trifoliate orange or its hybrids (7, 17) are almost certainly due to genetic incompatibilities. Other budunion problems have been attributed primarily to xyloporosis (9). Many others are of uncertain etiology (2, 5, 8, 13, 14, 15) and, in many cases, graft transmission has not been attempted. However, McClean (6) showed that a budunion crease of some sweet orange varieties on rough lemon rootstock was caused by a graft-transmissible agent. The budunion disorder of grapefruit on sour orange rootstock described by Reichert et al. (24) in Israel closely resembles the one described here. They did not attempt transmission, but indexing indicated that the disorder was not due to CTV. They also mentioned the possible involvement of xyloporosis in that budunion crease problem.

The affected trees in Texas were removed by growers after initial investigations were begun for fear of further spread. No new cases have appeared in the last 4 years. The occurrence of symptoms in young trees, many, if not all, of which came from the same nursery, suggests a problem derived from a budwood source and not from a naturally transmitted agent.
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