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Exocortis and Other Problems with Trifoliate Orange Rootstock

THE TRIFOLIATE ORANGE [*Poncirus trifoliata* (L.) Raf.], which is widely used as a rootstock in the region of Concordia, is afflicted by several abnormalities including exocortis-like stunting and scaling, stunting without scaling, scaling without stunting, and bud-union ring involving xyloporosis-like pits and death of trees when the Washington Navel variety of sweet orange [*Citrus sinensis* (L.) Osbeck] is used as a top. The result of some experimental work on these problems constitutes the subject matter of this paper.

Experimental Work

In October, 1956, Banfi and Beñatena established a plot for experimental transmission of exocortis. Marsh grapefruit (*C. paradisi* Macf.) buds were taken from a single tree that was suffering from exocortis and from a single tree that appeared to be healthy. These buds were grafted into trifoliate orange seedlings and each lot was set out in two rows of eight trees each, the two rows of infected trees alternating with the two rows of healthy trees. Two additional replications of this arrangement were made. A similar block of trees was started by grafting healthy buds from a Valencia sweet orange tree and a Valencia sweet orange tree affected by exocortis onto trifoliate orange seedlings.

During the first three years after the block was planted there was no difference in growth between the healthy and diseased trees. From the third to the fifth year, scaling developed in some of the trees and some of the trees became stunted. In the fifth year, the trunk circumference was measured both above and below the bud-union at a distance of 10 cm from the union. The average circumference for the healthy grapefruit stocks was 48.6 cm, for the scion 27.3 cm, for the diseased stock 30.9 cm, and for the scion 20.6 cm. For the healthy orange tree stocks the average circumference was 36.0 cm, for the scion 20.9 cm; for the diseased orange tree stocks 27.5 cm, and for the scion 18.0 cm.

The following conclusions were derived by observing the trees in the plot:

(a) Healthy trees made good development, had green foliage, and showed a rootstock conformation characteristic of healthy trees (Fig. 1,A); (b) Healthy trees were more or less of the same size, indicating little, if any, difference among seedlings of the trifoliate orange; (c) Buds from diseased trees gave rise to trees of variable reaction, some of them differing from healthy trees only by their apparently abnormal rootstock (Fig. 1,B), some being stunted, with and without scaling. The variations mentioned under (c) suggest the possibility of some buds being free of virus and others being infected with different strains of exocortis virus.

Distribution of virus in a single tree.—Budsticks were taken from a 12-year-old tree of Valencia sweet orange on trifoliate orange rootstock that was showing symptoms of exocortis. One budstick was taken from each of the following sectors of the tree: N, NE, E, SE, S, SW, W, and NW. Seven buds from each budstick were used to bud Rangpur lime (*C. limonia* Osbeck) seedlings. The results are summarized in Table 1. They demonstrate that there is great variation in the distribution of virus or virus strains in the tree. Plant number 2 inoculated with a bud from the NW sector remained healthy; plants 3, 4, and 5 had slight symptoms; plant number 6 had severe symptoms; and plant number 7 had moderate symptoms.

Plants in which the inoculating bud died did not develop symptoms, except plant 3 inoculated with a bud from the SW sector. Transmission of the virus of exocortis when the graft fails to take is apparently more difficult than in the case of psorosis virus, in which case almost all seedlings in which the inoculating buds did not take developed psorosis.

Transmission with different kinds of inoculum.—Rangpur lime seedlings inoculated by means of bark tissue from trees affected by exocortis have developed yellow blotches on the stem and splitting of the bark, proving that our lines of this variety are good indicator plants as are those in Brazil (6). From six to eight months after inoculation, these symptoms, and sometimes gum secretion and foliar deficiencies, are fully developed.

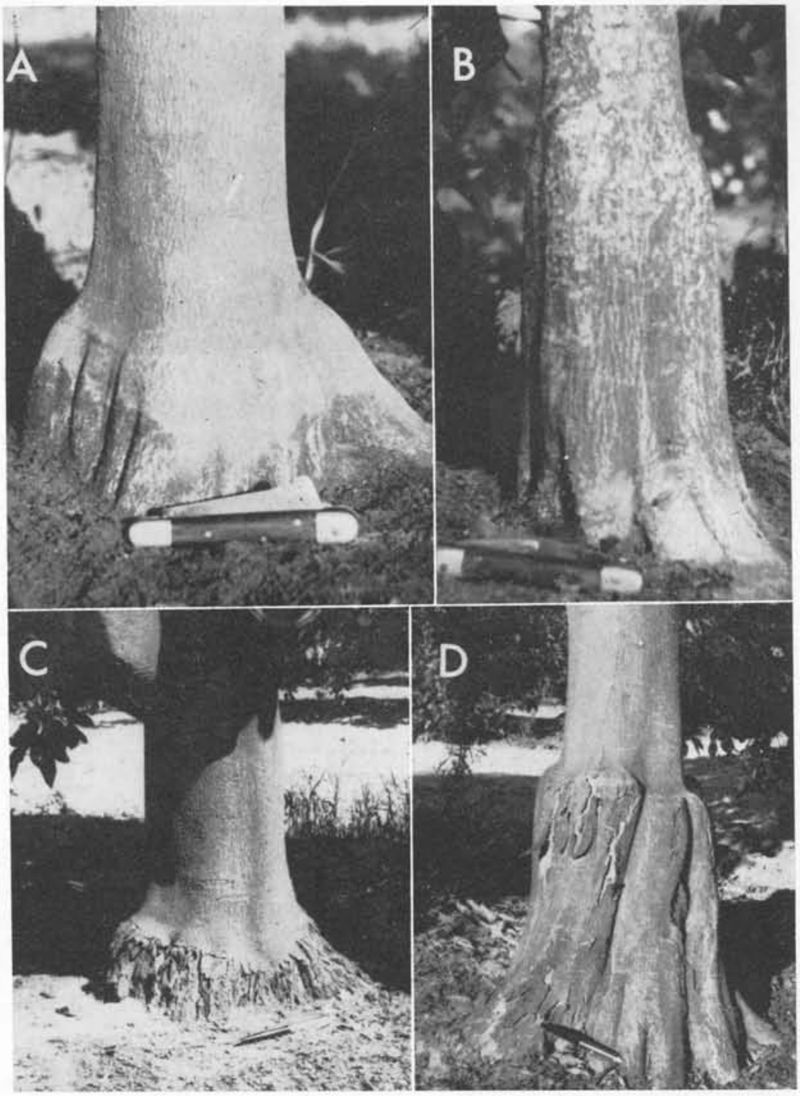

FIGURE 1. *A. Rootstock of a healthy grapefruit tree in the experimental plot; it is fluted and expands as it approaches the roots. B. Rootstock of an apparently healthy grapefruit tree in the block inoculated with exocortis virus; it shows a conformation different from that of a typical healthy tree. C. Trunk of a 15-year-old Marsh grapefruit tree on trifoliate orange rootstock; the tree has indexed negative in Rangpur lime seedlings but the rootstock is scaling. D. Trunk of a 20-year-old Valencia sweet orange tree on trifoliate orange rootstock; the tree is not stunted but the bark of the rootstock is beginning to scale.*

TABLE 1. Symptoms obtained in Rangpur lime seedlings after budding with buds from different sectors of a 12-year-old tree of Valencia sweet orange on a trifoliate orange rootstock that was showing exocortis-like scaling of the bark

	Number of Plant						
Sector	1	2	3	4	5	6	7
NW	–	0	1	1	1	4	3
NE	1	–	1	3	3	3	3
E	–	3	–	1	–	3	3
SE	3	–	–	–	3	1	–
S	2	2	1	2	1	2	2
SW	2	1	1(–)	1	4	1	2
W	2	–	–	1	–	3	3
N	1	2	1	–	1	1	1

– = no "take." 0 = without symptoms.
1 = bark lightly chlorotic. 2 = lightly chlorotic and with some splitting.
3 = moderate chlorosis and splitting. 4 = strong chlorosis and splitting.

When seedlings were inoculated by means of foliar tissue from a Marsh grapefruit tree on trifoliate orange rootstock showing symptoms of exocortis, there were no symptoms one year later; this result is comparable to that which Salibe (2) found. At the end of the second year, the seedlings showed yellow patches very similar to those seen in the first stages of exocortis. Eight months later, the yellow patches were abundant but there was no splitting of the bark.

Other disorders on trifoliate orange.—*Stunting*.—There are at least three kinds of stunting that occur in trees on trifoliate orange rootstock. One is found commonly in plots affected by exocortis and seems to be the same as that described by Fraser, Levitt, and Cox (3). This type is present in the experimental plot referred to above and is common in commercial plantations having a high incidence of exocortis. Some plants with this type of stunting show scaling of the rootstock and others do not. The rootstock generally has a prominent shoulder and its sides are vertical down as far as the crown roots.

The second type of stunting is shown by all Robertson Navel sweet orange trees inspected in the area. The rootstock does not have the conformation characteristic of the trifoliate orange. A line of gum is often deposited at the bud-union.

The third type of stunting is associated with decline and death of certain trees. It has been observed in a plot of the Washington Navel variety of sweet orange. Each year a half a dozen or more trees become stunted and develop a chlorosis at the time the fruit is ripening. Soon the leaves drop while the fruit remains attached to the tree; the fruit usually ripens earlier than those on a normal tree. In the inner part of the bark, there is a ridge that corresponds with a furrow on the outer bark and in which there is a deposit of gum. Starch is present only in the scion.

Scaling without stunting.—Many trees that show these symptoms (Fig. 1,C; 1,D) have been found. The scaling is similar to that caused by exocortis virus except that there is no stunting.

TYPE OF SYMPTOMS ON TRIFOLIATE ORANGE RELATED TO REACTION OF RANGPUR LIME.—Subinoculations were made by budding from ten of the trees on trifoliate orange rootstock to Rangpur lime seedlings, with the results indicated below.

(1, 2, 8) A ten-year-old Valencia sweet orange tree with typical symptoms of exocortis and a Villafranca lemon and a 20-year-old Valencia sweet orange tree showing scaling without stunting. A typical exocortis reaction was obtained on Rangpur lime; the seedlings developed chlorosis, splitting of the bark, and foliar symptoms of deficiency.

(4, 5, 7) A 20-year-old Valencia sweet orange tree beginning to scale (Fig. 1,D), a declining Washington Navel orange tree, and a stunted 15-year-old Marsh grapefruit having a definite shoulder on the rootstock. Symptoms in Rangpur lime seedlings were absent or mild; some seedlings had slight chlorosis and slight splitting of the bark.

(3, 6, 9, 10) A healthy Villafranca lemon tree, a healthy Washington Navel sweet orange tree, a 15-year-old Marsh grapefruit tree showing scaling without stunting (Fig. 1,C), and a healthy Marsh grapefruit tree. No symptoms in the Rangpur lime seedlings.

Discussion

When buds were taken from exocortis-affected trees in the experimental block and propagated in trifoliate orange seedlings, a variable reaction, from no symptoms at all to severe stunting and scaling, was obtained. Since the trifoliate orange seedlings were uniform, the variation must be attributed to presence of various strains of virus in the different buds and perhaps to the complete absence of virus from some buds. A similar hypothesis has been stated by Moreira (5), Fraser and Levitt (2), and Salibe (7). Results from subinoculation to Rangpur lime seedlings substantiate the hypothesis.

The failure to obtain early symptoms in Rangpur lime seedlings inoculated with leaf tissue from Marsh grapefruit trees affected by exocortis can be explained in one of three ways: (a) Exocortis virus was not carried in the leaf tissue; non-inoculated seedlings did not, however, develop yellow patches similar to those that developed after about a year in the inoculated seedlings. (b) There is a lower concentration of virus in leaf tissue than in buds and a longer time is therefore required for severe symptoms to be induced in the seedling. (c) There is a tendency for mild strains of exocortis virus to dominate in leaf tissue. Grant (4) reported that inoculation with leaf tissue of Key lime plants carrying a mild strain of tristeza virus usually led to mild symptoms in the inoculated plant whereas the symptoms were variable when inoculations were made with leaf tissue of plants carrying more severe strains, suggesting that there is a tendency for the mild strain to be dominant in leaf tissue.

Subinoculations to Rangpur lime seedlings from trees showing scaling without stunting have given two types of reactions: (a) a positive exocortis response and (b) a negative response. We can suppose that (a) the rootstock of the source tree is genetically resistant, (b) a mild strain of virus was present, although a mild strain should cause some stunting, (c) the tree was not propagated from infected budwood but was subsequently infected by means of a vector, or (d) stunting and scaling result from a complex that occasionally separates into two or more components. Further work is required to decide among these various possibilities.

Literature Cited

1. Banfi, A., and Beñatena, H. N. 1956. El pie de trifolio. Actualización y observaciones sobre su comportamiento en la zona de Concordia. Nuevos ensayos iniciados. IDIA 101: 37-42.
2. Fraser, L. R., and Levitt, E. C. 1959. Recent advances in the study of exocortis (scaly butt) in Australia, p. 129-133. *In* J. M. Wallace [ed.], Citrus Virus Diseases. Univ. Calif. Div. Agr. Sci., Berkeley.
3. Fraser, L. R., Levitt, E. C., and Cox, J. 1961. Relationship between exocortis and stunting of citrus varieties on Poncirus trifoliata rootstock, p. 34-39. *In* W. C. Price [ed.], Proc. 2nd Conf. Intern. Organization Citrus Virol. Univ. Florida Press, Gainesville.
4. Grant, T. J. 1959. Tristeza virus strains in relation to different citrus species used as test plants. Phytopathology 49: 823-827.
5. Moreira, S. 1959. Rangpur lime disease and its relationship to exocortis, p. 135-140. *In* J. M. Wallace [ed.], Citrus Virus Diseases. Univ. Calif. Div. Agr. Sci., Berkeley.
6. Moreira, S. 1961. A quick field test for exocortis, p. 40-42. *In* W. C. Price [ed.], Proc. 2nd Conf. Intern. Organization Citrus Virol. Univ. Florida Press, Gainesville.
7. Salibe, A. A. 1961. Contribuição ao estudo da doenca exocorte dos citros. 71 p. Mimeographed. Doctorate Thesis. Univ. de São Paulo.