DEVELOPMENT AND MAINTENANCE OF VIRUS-FREE PROPAGATING MATERIAL

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Plants that have been propagated vegetatively for many years are usually infected with one or more viruses. This is a principle of plant pathology. Not all viruses cause conspicuous disease symptoms in plants they infect; some may not cause any detectable damage and in some cases they may perhaps even be desired by man. Examples of the use of viruses, often unknowingly, to make desired modifications of plant growth are certain variegations in ornamentals, shortened internodes and dwarfing in some fruit-tree varieties, and recumbent or weeping growth habits of some woody ornamentals. In most cases, however, viruses are harmful to plants, and clean stock is preferable to diseased stock.

The need for virus-clean stock.—When damage caused by viruses is not conspicuous, losses are often difficult to ascertain. A moderate reduction in fruit set, fruit size, or tree or vine size may easily go unnoticed or be attributed to cultural or environmental factors, especially if all plants in a planting are infected uniformly. Losses can be measured accurately only by comparing infected plants with known clean plants under similar conditions. Not much work has been done along this line by plant pathologists or horticulturists. What has been done has paid good dividends by encouraging the use of virus-clean stock. A recent report[1] from England attributes to virus infection a yield depression of 30% in sweet cherries. In California, annual losses due to virus and virus-like diseases are conservatively estimated at close to $1,000,000 in cherries, more than $2,000,000 in peaches, and over $10,000,000 in grapes. Most of these losses could have been prevented if virus-clean stock had been available and used for planting the orchards and vineyards presently in use.

Virus-clean stock of many varieties of fruits and grapes and some ornamentals is available[2], and more is becoming available each year. The rate at which additional varieties are cleaned up will depend more or less on the desire of nurserymen and growers for such stock and on the performance of virus-clean stock in commercial plantings.

Methods of obtaining virus-clean stock.—Plant propagators, by carefully selecting the very best looking and best performing plants for propagating new plants, may, knowingly or unknowingly, avoid many viruses. This visual selection process can be helped by recognizing virus symptoms. Even so, some viruses may not cause obvious symptoms, so indexing

[1]Report, Committee on Crop Losses in California, 1965
[2]California Foundation Plant Materials Service, University of California, Davis, and IR-2 Repository for fruit trees, Prosser, Washington.

must be used to detect infection. In indexing, symptomless or latent viruses are detected and identified through the use of plant species and varieties specially selected because they show diagnostic symptoms of known virus diseases.

Indexing with plant hosts.—Some viruses can be transmitted by juice to index plants, which is the simplest indexing procedure and the easiest to apply. A small amount of suspect young leaf or flower tissue, or sometimes root tissue, is macerated by mortar and pestle in a suitable buffer and preservative chemicals. The triturated material is gently rubbed on the cotyledons or leaves of selected indicator plants that are first dusted with carborundum. Inoculated surfaces may be washed immediately with water, or simply blotted dry with absorbent paper. The plants are then observed for a few days to several weeks for indications of virus infection. Favorite choices for indicators are common plants such as tobacco, bean, cucumber, squash, pigweed, and cowpea. The species that become infected and the symptoms they show help to identify the virus. Cucurbit and pigweed species have become standard hosts for detecting several different viruses in stone fruits and apple and pear. Mechanically transmissible viruses in herbaceous ornamentals also are easily detected with one or more of the plants mentioned above.

Grafting to suitable indicators can detect viruses that are not mechanically transmissible or not easily recovered from host plants that have inhibitors. Grafting can be done between herbaceous as well as between herbaceous and woody plants. We routinely make successful grafts for virus transmission between pigweed and apple and cherry. From these hosts we can easily transmit virus by tissue grafting to any rosaceous plants. Virus may go from either host into the other.

Grafting is usually used to transmit virus from donor woody plants to other woody plants chosen for their ability to detect and identify viruses. Thus, for the stone fruits almost all known viruses can be detected by visual inspection combined with indexing on 'Elberta' peach, 'Bing' cherry, 'Tilton' apricot, 'Montmorency' sour cherry, and 'Shiro-fugen' flowering cherry. For apples the known viruses can be detected by visual inspection plus indexing on about 8 selected indicators. Similarly, minimum indicator hosts are presently being selected for pear and woody ornamentals. Unfortunately, tests on woody indicators require several months to several years, so plant pathologists are constantly looking for faster methods.

Indexing by chemical and physical methods.—Several chemical and physical methods of detecting virus infection are known. Only a few have proved practical and reliable, however. There is promise in this area of research, though, and if enough effort is made I am sure practical and reliable tests could be developed.

Curing virus-infected plants.—Not infrequently selection and indexing reveal that no virus-clean plants are available in

a particular variety. Then it becomes necessary to produce clean plants from the infected ones. This is usually done by heat therapy, by tip or meristem culture, or by a combination of these methods. Most viruses do not infect the seeds of their host plants. In such cases, clean seedlings can be obtained from infected plants. For clonally propagated plants this is of little significance except for rootstocks.

Heat therapy to obtain virus-clean clones has proved of great practical value with fruit trees, grapes, and some ornamentals. When plants in containers are exposed continuously for 3-12 weeks to dry heat at 100°F, the viruses are inactivated in at least portions of the tops. Cuttings or buds can then be removed and used to produce new plants which are free of virus. Not all plants propagated from treated plants are necessarily virus-clean, however, so post-treatment indexing is necessary in most cases.

Tip or meristem culture has been used successfully for obtaining virus-clean plants of several species, such as Irish potatoes, sweet potatoes, carnations, and pome fruits. The same techniques are used to obtain clean plants of some species from plants that are infected systemically with fungi or bacteria.

Maintaining virus-free propagating material — Foundation stock is developed from individual plants that are horticulturally superior and show no evidence of virus infection in visual inspection or indexing. The procedures chosen to maintain clean stocks may vary somewhat by crop and location. Most important is that the procedures must be based on certain basic principles that assure success in a clean-stock program. Short-cuts may be all right as long as they do not violate principles.

The principles on which clean-stock programs must be based are as follows:

1. Foundation or "nuclear" stock must be horticulturally desirable, inspected visually for virus and genetic disorders, and indexed by proved methods that give reliable results.
2. Foundation or "nuclear" (first-stage) stock must be protected by natural or artificial means against infection.
3. Understocks of a quality comparable to that of topstocks must be available.
4. Mother blocks, scion orchards, or second-stage propagation sources must be propagated by cuttings directly from foundation stock, or by grafting on virus-clean rootstocks if no indexing is done before further increase.
5. Second— or third-stage propagation sources, sometimes called "increase plantings," can be used for only limited times without reindexing and must be replaced periodically from first— or second-stage stock.
6. Regular inspection of all propagation sources by qualified inspectors is necessary if certification is to be meaning-

ful. Certification should be for procedures rather than for virus-cleanliness.

A minimum clean-stock program, then, would consist of: (1) a foundation block of plants true to variety, horticulturally desirable, and free of viruses and genetic disorders; (2) mother blocks or second-stage plantings conveniently located for increase of budwood, and regularly inspected and sample-indexed; and (3) rootstocks that meet standards comparable to those for topstock, or with acceptable variance.

LITERATURE CITED

1 Posnette, A F 1963. Virus diseases of apple and pears Tech Comm. #30 Commonwealth Bur Hort and Plant Crops East Malling

2 Posnette, A F, R Cropley and A A J Swait 1968. The incidence of virus diseases in English sweet-cherry orchards and their effect on yield' *Ann Appl Biol* 61·351-360

MODERATOR RODEBAUGH: Thank you George. Our next speaker on the program today is Mr. Stan Mather who is Chief of the Nursery Service Division, California Department of Agriculture. Stan will give us a brief review of the certification program and then we will see a film which describes the program in a little greater detail.

CALIFORNIA'S NURSERY STOCK REGISTRATION AND CERTIFICATION PROGRAMS

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Nursery stock is a primary means by which plant pests may be introduced. Even though recognized as such and given close attention over the years, serious virus diseases and other pests have been perpetuated unwittingly by man through his propagation of plants. The exclusion of plant pests from nursery stock propagating sources is not an easy task. Close coordination between research, regulatory and industry activities is needed if we are to succeed. Index testing for virus diseases, laboratory techniques for the detection of nematodes, and eradicative treatments have and are being developed. These improved procedures will permit nurserymen to maintain plant propagative sources for the production of high quality nursery stock.

Regulatory and industry activities in past years have been directed toward reducing pest infestation through visual observation of plants and by quarantine restrictions. Experience has shown that certain plant pests can be present that were either not recognizable or not detectable by ordinary means of inspection. Emphasis has shifted to inspection of nursery stock at origin and exclusion of plant pests from nursery grow-