SOURCE SELECTION OF VEGETATIVELY PROPAGATED CULTIVARS

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The *clone* is a primary concept in horticulture, with vegetative propagation (cloning) being one of our most important procedures (4,6). Most cultivars propagated by nurseries today are clones. Success in propagation depends upon maintaining trueness-to-cultivar and trueness-to-type. Research in this area, however, has emphasized the detection and elimination of viruses in clonal materials (2,10,11).

The purpose of this paper is to discuss some basic concepts of source selection from a genetic standpoint and to describe some unique problems with clonal variability. Emphasis will be given to the propagation of fruit and nut crops, particularly California almonds.

Most clones originate as selections of superior individuals from seedling populations. Selections are vegetatively propagated for test plantings. Figure 8-17 in Hartmann, et al. (4) outlines the basic steps in the development and subsequent propagation sequences. An alternative approach is selection of bud-sports (bud-mutations) from within the clone. All but a few almond cultivars originated by the selection of individual plants from seedling populations. Some, including 'Nonpareil', 'Mission', 'Ne Plus Ultra', 'Peerless', 'Drake', and 'I.X.L.', originated over a hundred years ago and have a long history of consecutive generations of propagations. Others, such as 'Merced', 'Carmel', and 'Thompson', originated more recently, either from chance seedlings in commercial orchards, roadsides, etc., or from public and private breeding programs.

Individual trees of the clone are selected to be the *source* to begin a sequence of propagation. Most commercial propagation of fruit and nut trees in California is by T-budding cultivar material to seedling or clonal rootstocks. Individual vegetative buds are taken from a budstick of 8 to 10 buds. The related lines of descent through propagations from the original selection constitute the *pedigree* of the cultivar. In some of the younger almond cultivars, the original trees and representatives of all the intervening generations of the propagation sequence are in existence.

PEDIGREE SYSTEM OF SELECTION

Most commercial nurseries for fruit and nut trees in California have utilized the *pedigree system.* Commercial orchards are used as *source blocks* for collecting propagation material. Budwood is collected from bearing trees after *visual inspection*, avoiding individual trees which do not appear *true-to-type.* Identity with the source orchard, and often with the individual source trees, are maintained into the nursery row. When the nursery trees are dug in the fall, the trees are graded by size and any relationship to specific source trees, or even orchards, may be lost.

The same orchard source may be used for a number of years until the trees lose vigor and produce reduced amounts of suitable budwood. New budwood sources are then chosen which are invariably the vegetative progeny of previously used orchards. As this: source > > nursery > > progeny orchard cycle is repeated, a series of vegetative generations of individual ''budlines'' evolve from separate nurseries, each with a unique propagation history or pedigree.

NUCLEAR STOCK (OR SOURCE-CLONE) SYSTEMS

This system starts with a single source plant (or even a bud) which then provides a ''nucleus'' for all subsequent propagations (2,3). This plant essentially creates a ''new'' clone selected from the original clone. The primary reason for this procedure has been to clone a source plant found to be free of specific viruses. It is assumed that the new plants will have the same genotype as the original clone. (Use of the term ''source-clone'' distinguishes these special kinds of ''clones'' from traditional new clones that are chosen because of a mutation that actually changes the genotype.)

The *source-clone* is subsequently maintained in a special *scion orchard* for budwood production rather than for a crop. Trees usually need to be multiplied in an *increase block* in order to provide sufficient volume of budwood for nursery propagation.

Nuclear Stock System

In the mid-1960's a nuclear stock system involving *Registration* and *Certification* was introduced by the California Department of Food and Agriculture to distribute selected indexed source-clones of grape, fruit, and nut tree species. There are three general phases (4).

Phase I. Selection of specific source-clones which had been indexed, found to be free of specific viruses, and appeared to be true-to-type.

Phase II. Maintenance of trees (2 or more) in a *Foundation Orchard* under protected conditions to prevent reinfection and to retain their genetic identity. This function was carried out by the *Foundation Seed and Plant Materials Service (FSPMS)* of the University of California.

Phase III. Distribution of ''registered budwood'' to commercial nurseries who would establish scion orchards and produce ''certified nursery stock'' under the regulations and supervision of the California Department of Food and Agriculture.

Some attempts were made at the time by the commercial fruit and nut tree industry to adopt this program but very little material was actually distributed and the program was poorly utilized. Several reasons can be cited:

a. Recovery of the added cost of the operation in the sale price of the tree was not possible.
b. Inspections and cumbersome management practices were required.
c. Some nurseries had more confidence in their own material obtained by pedigree selection,
d. Problems arose with genetic disorders, trueness-to-type, and knowledge of horticultural quality, which reduced industry confidence in not only specific source-clones but also in the program.
e. Many important cultivars, particularly those newly patented by commercial firms were not included in the program.

Scion Orchard Production Systems

Consequently, some commercial nurseries began to incorporate virus testing into their own programs and to develop scion orchards to manage their own sources. Registered source-clones were sometimes used but other sources were also included, such as privately patented cultivars. These sources often did not have the level of virus testing to which registered stock was subjected.

In the past few years, there has been renewed interest by commercial nurserymen in California to reestablish a Registration and Certification program for fruit and nut crops as a basic tool for nursery source management. This trend has been fostered by the following reasons:

a. Problems arose with virus infections and associated threats of litigation.
b. A program to provide financial support for a ''clean stock'' program from fees on commercial nursery stock was instigated.

c. Competition developed from certified stock production from other states.
d. There was improved understanding of genetic problems within clones leading to potentially more effective practices for maintaining trueness-to-type (1,7).

SOME BASIC PRINCIPLES OF SOURCE SELECTION

The model: PHENOTYPE = GENOTYPE + ENVIRONMENT states that the appearance and performance of any individual plant is a function of its genes interacting with the environment. What we perceive is the *phenotype*. What is propagated is the *genotype*. A high correlation exists in clones between the genotype and associated phenotype. This means that the phenotype of the offspring should be very similar to the phenotype of the source plant since their genotypes are identical.

Two basic terms have been used to describe phenotypes in relation to source selection:

Trueness-to-cultivar means that a specific identified cultivar is being reproduced and not some other cultivar that has been propagated by error.

Trueness-to-type means that not only is the correct cultivar being introduced but also that the plants being produced are typical for that cultivar and meet standards for performance and appearance. In practice, the two terms are often used interchangeably but the two are fundamentally different.

Trueness-to-type applies both to *source* plants and to *vegetative progeny* plants. If the two are different, then change has occurred in the cultivar, (a) in the source plant (or its antecedents), (b) during the propagation process, or (c) in the vegetative progeny. In dealing with deviations from ''trueness-to-type'', one first needs to be able to determine the cause of the variation and then to identify the exact time in the propagation sequence when the variant appeared. Variation may result from three basic causes: pathogen-induced (primarily viruses), genetic changes, or environmental effects. In addition, there may be unique *species or cultivar-specific genetic disorders*, as described later in this paper.

Three standard procedures are used to determine the causes of variation:

Virus Indexing. This procedure includes various types of tests, including transmission, biochemical, and other procedures, and provides direct evidence for the presence of specific systemic viruses within individual plants (8,10). A full range of tests are specified in the Registration and Certification regulations for fruit and nut trees in California. These tests provide a category of *indexed* source plants that are available for propagation. In our

model, the virus combines with the environment to affect the phenotype expression:

phenotype = genotype + (virus + environment)

The effect on the phenotype varies by virus, cultivar, and environment. Sometimes the combination can kill plants. In other combinations, the effect may be severe and result in decline, yield reduction, or poor quality. However, in some combinations, the phenotypic effect is not readily apparent and not easily measured.

Nevertheless, the trend is for the elimination of identified viruses from propagation stock when it is possible. Plant pathologists and propagators, however, are careful not to use the term ''virus-free'' in referring to such plants. The term should only apply towards those viruses for which the test has been made. A more appropriate term is ''indexed-stock'' which refers to source plants in which the absence of specific viruses and other systemic pathogens has been verified by specific indexing methods.

Phenotypic Selection. This term refers to selection of trueness-to-type by VISUAL INSPECTION of the source plants. Most organisms have species- or cultivar-specific characteristics that an experienced evaluator can recognize, providing that the inspection is made under proper environment and management conditions. A high correlation between the morphological characteristics of a source plant and its vegetative progeny is expected. Consequently, visual inspection of the source plants becomes an essential part of any selection process and, for some plant cultivars, may be the only procedure available. The procedure can be effective, providing that the inspection is carried out under the right conditions, done by knowledgeable persons and utilizes specific standards of identification.

Phenotypic selection has important limitations, however. Some traits—e.g., health, yield potential, and vigor—may not be readily evaluated or can be strongly influenced by the environment, age, location, or management of the source plant. Fruit and nut trees grown for budwood are pruned severely to increase new growth and reduce flowering. Under these conditions, fruit and nut characteristics invariably are atypical and observations may be misleading. Visual inspections may thus result in ''false readings'' and misdiagnosis of genetic ''problems.''

Visual inspections may not detect viruses, latent mutations, yield reduction, or susceptibility to latent disorders, such as *noninfectious bud-failure* in almonds.

Genotype selection. This procedure is based on *visual inspection* of the *vegetative progeny* and provides a test for the actual type of plants that the source will produce. Important uses of a vegetative progeny test include:

a. the screening of sources for latent genetic mutations, such as nonproductive syndrome in almond, and other disorders, including noninfectious bud-failure (almond), crinkle (cherry), etc.
b. the testing of yield performance and horticultural quality.
c. establishing whether a perceived abnormality is due to environmental or genetic causes.

For these reasons, genotypic selection should be incorporated into source selection programs whenever possible.

SPECIFIC PROBLEMS AFFECTING SOURCE SELECTION IN ALMOND

This section illustrates the concepts and application of source selection by describing experiences we have had with the almond.

1. Virus problems. The almond is susceptible to the same range of viruses affecting other stone fruit species. General procedures for viral elimination and maintenance of source materials are available. Most almond cultivars do not appear to have as great a phenotypic response to some common viruses as other stone fruits. Nevertheless, the use of "clean" material is desirable.

2. Genetic problems: mutations, chimeras, budsports. These terms refer to discrete changes in the genes, chromosomes, or other genetic units in single cells somewhere in a growing point (4). Normally mutations are rare and the probability of one occurring in a selected source plant is low. Mutations in a cell in a growing point may lead to a *chimera*, which may then encompass a sizable part of a plant and affect a number of buds. The chimeric shoot may be latent and not easily identified by visual inspection. If such an undetected chimeric shoot is used as a source of single bud propagules, the probability of off-type progeny plants being produced is high.

This type of single mutation will be referred to as Type I. In the 1950's, a late-blooming mutant of the 'Nonpareil' cultivar was discovered which involved several entire trees and single limbs of others (8). Evidently a single gene mutation had occurred but was not detectable until after propagation. The mutant became the patented cultivar, 'Tardy Nonpareil'. In this case, the genetic change affected few trees, and proved economically useful. In another example, a heavy producing 'Nonpareil' limb was discovered which became the patented 'Jeffries'. It was later found that the pollination requirements of the budsport were unique (9) and resulted in economic problems when planted in commercial orchards.

Nonproductive syndrome or "bull" trees (a second type of mutation problem—referred to as Type II) reduced productivity,

increased vigor, and produced morphological aberrations of the fruit and foliage (7). Trees with this condition began to appear in commercial orchards in the late 1960's and early 1970's and were associated with specific nursery sources and cultivars. After considerable research, we were able to trace the origin of the problem through several vegetative generations to two nursery sources. Our research did not indicate a pathogen problem but rather some earlier "event" which resulted in an "explosion" of mutations that appear to affect a number of traits. The incidence of these characteristics was associated with exposure to certain agricultural chemicals, although this relationship has not been experimentally tested. The key point is that this problem was not detected in the primary source plants where it was initiated but only in secondary progeny plants after several generations of consecutive propagation.

3. Species or cultivar-specific "genetic disorders." In almond a genetic disorder called *noninfectious bud-failure* (BF) falls into this category (5,9). This condition does not have a known virus etiology but develops in a regular pattern within specific cultivars (1). Its primary action involves the necrosis of vegetative buds and consequently dieback, which after consecutive years, results in a BF-induced phenotype called "crazy-top." Physiologically, the disorder appears to involve the loss of resistance to stress (high temperatures, low moisture) with prior growth required for expression. Variation in the potential for BF is shown by different propagation sources. Thus, the primary method of control has been through source selection, though subsequent conditions at the progeny orchard site may also be important. Selection cannot be based upon the phenotypic selection of the source but requires information from progeny testing.

4. Nongenetic causes. These produce only temporary effects and are not heritable. These can lead to false readings. For example, in the investigations on the *nonproductive syndrome*, source trees growing in a scion orchard, which were heavily pruned for budwood, showed low crops, extra vigor, and abnormal nut morphology. Visual inspection of these plants led to the misdiagnosis of a Type II disorder. This error was only revealed by conducting vegetative progeny tests of the sources involved.

SUMMARY AND CONCLUSIONS

In the selection of propagation sources for vegetative propagation, testing for viruses, trueness-to-cultivar, and trueness-to-type are required. These tests are particularly important for source-clones since all subsequent propagules will inherit identical

genetic factors. Our studies indicate that visual inspections under proper growing conditions are essential. Visual inspections of the source plants (phenotypic selection), however, are not always adequate. Additional visual inspections of the progeny plants (genotypic selection) are necessary particularly when dealing with latent disorders.

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