# Alternatives to rootstock propagation by seed: differences, concerns and expectations

#### By Ute Albrecht

ommercial citrus rootstock propagation traditionally occurs via use of nucellar apomictic seed, which generates genetically identical seedlings that are used as liners for budding. With the high demand for superior rootstocks in the age of HLB, commercial nurseries are currently experiencing a shortage of seeds for some of the most popular rootstock varieties. In addition, for many of the newer rootstocks developed by the University of Florida Institute of Food and Agricultural Sciences (UF/IFAS) and the U.S. Department of Agriculture (USDA) citrus breeding programs, no seed source trees are yet available.

As a consequence, rootstock propagation has to rely on alternative methods such as cuttings or tissue culture (TC) to generate the large number of rootstock liners needed by nurseries. Like plants derived from apomictic seed, both of these alternative propagation methods will produce genetically uniform plants.

# CUTTINGS

For cuttings propagation of rootstocks, single-node stem cuttings (about 1 inch in length) are often used. Cuttings are removed from recently matured branches of citrus plants that must be certified disease-free. Leaves should remain attached to the node to allow photosynthates to be delivered to the developing roots but can be reduced in size (Figure 1A).

The basal end of the cutting is usually dipped into a powder or other material that contains root-stimulating hormones and then inserted into the potting medium. Under high-moisture conditions, a young plant will develop within a few weeks from the node, and roots will begin to form.

# **TISSUE CULTURE**

The starting material for TC propagation (micropropagation) depends on the preference of the nursery and can consist of nucellar embryos, buds or other tissue. All starting material must be derived from certified disease-free plants of true-to-type material of the desired rootstock. Typically, the starting material will come directly from the Florida Department of Agriculture and Consumer Services Division



**Figure 1.** A) Single-node cuttings inserted into potting mix. B) Tissue culture (TC)-generated shoot clusters (photo by Beth Lamb). C) Root growth of TC-generated young rootstock plant.

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The explant (the extracted plant organ or tissue piece) is placed into an agar-nutrient medium to generate clusters of multiple shoots (Figure 1B). This process occurs under sterile conditions. Shoots are then separated and either rooted on agar medium before transplanting or directly placed into potting medium where roots will form within a few weeks (Figure 1C).

Care of young rootstock plants generated from cuttings or TC then continues in the same manner as for seedlings. TC propagation has many advantages: it provides pathogen-free plants, it allows the rapid propagation of large numbers of very uniform identical plants, and it can be done without seasonal restrictions. It is a major propagation tool for the commercial production of many fruit tree rootstocks other than citrus.

Despite many advantages of TC propagation, a survey conducted in 2017 indicated that the majority of citrus nurseries in Florida prefer rootstock liners that originate from seed over those that originate from TC. In addition to the anticipated higher costs of plants from vegetative origin, there is concern these trees will have an inferior root system and inconsistent vigor. The nurseries conceded, however, that continued improvement of TC techniques will likely improve the quality of liners they receive.

Besides nursery-related issues, there are also concerns that trees on rootstocks not produced from seed will be more susceptible to strong winds or hurricanes. The recent experience of Hurricane Irma, which uprooted many citrus trees along its path across Florida's production areas, has likely amplified these concerns.

# ROOT COMPARISON RESEARCH

How different are the root systems of plants propagated through cuttings or TC compared with seedlings? Generally, a plant grown from seed will have a single and well-defined taproot from which multiple smaller roots arise (Figure 2A). In contrast, TC or cuttings propagation will generate a root system that is composed of multiple adventitious or lateral roots instead of a taproot (Figure 2B). Although a few studies were conducted in the 1950s and 1980s that compared growth of seedlings and rooted cuttings, we do not know much about the short and long-term effects of propagation method, especially TC, on grafted citrus trees under present commercial conditions.

In order to provide this much needed information, we have initiated a collaborative study with Kim Bowman (USDA, Agricultural Research Service, Fort Pierce), Beth Lamb and Phil Rucks (Phillip Rucks Citrus Nursery, Frostproof), Mireira Bordas (Agromillora, Wildwood) and Anna and Nate Jameson (Brite Leaf Citrus Nursery, Lake Panasoffkee) to propagate different rootstocks by seed, cuttings and TC. The rootstocks included in this study are Swingle, Cleopatra, US-802, US-812, US-897, US-942 and X-639. All of these are among the top 10 rootstocks propagated in Florida in recent years, according to the Citrus Budwood Annual Report of 2016.

Initial observations on a subset of young rootstock plants during the early nursery stage showed considerable



**Figure 3.** Pushed citrus trees growing on the Central Florida Ridge. Note the absence of a deep tap root. The rootstock is Carrizo.

differences in root structure and other plant traits based on propagation method. As expected, plants propagated by seed generated mostly one well-defined taproot, whereas plants propagated by cuttings and TC generated multiple adventitious roots (Figure 2). Although the root mass fraction was larger in seedlings, root length of fibrous roots per unit root mass was larger in cuttings and TC plants. We concluded that the plants generated by cuttings and TC are very efficient in their ability to uptake water and nutrients at this early stage of development. This suggests that commercial nurseries will likely have to adjust their management practices based on the method by which rootstock liners are produced.

Another important observation

from this study was that root structure varied considerably among the different rootstock varieties, no matter the propagation method. Differences were most prominent between the low-vigorinducing rootstock US-897 and the highvigor-inducing rootstock US-802.

We continued our study to analyze root structure and plant traits in grafted field-ready Valencia trees. Interestingly, we found little difference



**Figure 2.** Typical seedling root system (A) and cutting-derived adventitious root system (B) of young citrus rootstock plants.

for most plant growth variables that were associated with rootstock propagation method. Unlike what we observed at the early growth stage of rootstock liners, the root mass fraction of the grafted field-ready trees was not different between trees on seed-propagated rootstocks, cuttings or TC-propagated rootstocks. But, for some rootstocks propagated by seed, the percentage of larger-diameter roots was higher.

# WITHSTANDING WIND

What does this research mean for a citrus tree's ability to withstand strong winds? It is commonly thought that trees of vegetative origin are prone to wind-induced uprooting because of the absence of a taproot. In the heavy flatwood soils on the east coast and the southwest Florida citrus-growing regions, however, taproots generally do not extend deeply into the soil. In present day, the same has been observed for citrus trees grown in the well-drained sandy soils in the central growing regions of Florida (Figure 3, page 10).

One reason for absence of a deep taproot on large trees, even in deep sandy soils, may be the switch from overhead to micro-irrigation. Another reason may be the cessation of taproot growth in the confined environment of the growing container in the nursery that we observed in our studies.

It is apparent that under the current citrus-production conditions, root architecture and anchorage in the upper zone of the soil are the most critical factor impacting the susceptibility of trees to wind-induced uprooting. An important finding of our studies was the notable difference of root architecture among rootstock varieties independent of the propagation method. These differences were the same in grafted trees and in the ungrafted rootstock liners. Therefore, we expect that rootstock-specific traits will have a larger influence on field performance of a commercial citrus tree than the method by which it was propagated.

More than 400 trees from this study were field planted last year at the UF/IFAS Southwest Florida Research and Education Center in Immokalee. More field trials were planted this spring in two commercial locations in collaboration with Larry Black (Peace River Packing Company) and Joby Sherrod (Duda Fresh Food and Farms). Short- and long-term evaluation of these field trials will hopefully resolve concerns about the quality of citrus trees propagated by methods other than by seed.

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# Definitions

**Apomictic** – reproduced asexually (without fertilization)

**Nucellar embryony** – a form of seed reproduction in many citrus varieties in which genetically identical embryos develop from the nucellar tissue

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