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## SUBTROPICAL FRUIT TREE PRODUCTION: AVOCADO AS A CASE STUDY

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**Abstract.** A general description of techniques in a modern commercial avocado tree nursery and a summary of special precautions for the prevention of infections by *Phytophthora* organisms, sunblotch virus, and *Rhizoctonia* are given. Also, a description of special procedures for seed storage and a recently patented method for producing grafted avocados on clonal rootstocks is described.

## INTRODUCTION

I have chosen avocados to discuss because, frankly, the avocado (*Persea americana*), is one of the few plants I know any-

thing about! It is suitable as an example of subtropical horticulture for it is evergreen, frost-tender, grows well on acid soils, and is native to areas with frequent summer rains. It is a difficult plant to grow really well in California and we of the avocado nursery industry use a number of specialized techniques to achieve efficient production. One of the rewarding aspects of our work is that we produce plants for a sophisticated commercial clientele whose interest is in premium, durable, and disease-free trees which are planted in highly capitalized orchards for commercial fruit production.

Radical changes have occurred in California avocado nurseries during the past 25 years. Twenty years ago most plants were started in fields in nursery rows, budded in place, and balled in burlap. The process required from 3 to 4 years and the percentage of successfully budded trees that resulted was very low by today's standards. The first really radical commercial departure from this traditional procedure is attributed to Walter Beck, one of the pioneers and early masters of avocado propagation. Beck began propagating under greenhouse conditions and was able to produce small and tender, but healthy, containerized plants, with a then high percentage of success, in only 8 months (1). He accomplished this by utilizing bottomless cylindrical containers of building paper, which measured 5" (12.5cm) in diameter by 12" (30.5cm) tall, and by tip grafting small seedlings. Tip grafting was simply a form of whip grafting, using apical sections of stems of winter wood as scion material.

Today's standard techniques are a blend of traditional methods with techniques worked out by Beck. We utilize Beck's greenhouse discoveries to assure ourselves of reasonable propagational efficiency; we finish our trees off in the open sun to achieve sturdiness which is almost essential when the trees are transferred and planted in their permanent orchard locations, sometimes under hot and arid conditions.

## BASIC GROWING PROCEDURES

At Brokaw Nursery we start most of our seeds during the fall as they are harvested. Seeds of the Mexican race are chosen because, on the average, they are most suited to our California soil and climate. We scarify the seeds by slicing thin sections from the basal and apical ends, and bed them in a peat moss-perlite blend until they germinate and demonstrate sound, healthy, seed tissue and roots. Then we transfer them to gusseted and perforated plastic bags which measure about 2-1/2" (5.3cm) in diameter by 9" (23cm) in length. The bags have been previously filled with the peat moss-perlite blend. This type of

packaging furnishes us with a very concentrated grouping of small seedlings which reach grafting stage about 6 weeks after scarification and bedding of the seeds.

For grafting we depart from Beck's whiplike procedures, for our propagators can work most efficiently with less exacting techniques. We first cut off the seedling squarely, about 4 in. above the bag. Then we cut a vertical slit in the small stock (now about 3/16" to 1/4" (.5-.65cm) in diameter) and insert a scion, the butt end of which has been sliced on two opposite sides to form a wedge at the points of cambial contact. The diameter of the scion is relatively unimportant as we merely slice off one edge if it is oversized, so as to make an acceptably neat juncture. The area of union is then wrapped with rubber, and asphalt grafting emulsion is applied to the wounds. Some of our grafters have reached high success rates in this work. We employ ladies who graft close to 100 per hour much of the time, and achieve success in a good 90% of their production.

Assuming that one chooses suitable graftwood, and handles it properly, dessication is the greatest hazard for successful "take" of avocado grafts. We avoid this problem by watering our plants just before grafting and placing them in special high humidity chambers immediately following grafting. They remain in the chambers for about 10 days. We believe that this procedure produces more uniform graft development, as well as increased success in grafting yields.

After removal from the humidity chambers, the young grafts are transferred to standard greenhouse conditions for a period of approximately three weeks. They are then sorted for size, transferred to benches under 50% shade for at least 5 days, and subsequently transferred to open sun pallets, and then to the field containers any time after 10 days from leaving the greenhouse.

A good aspect of this procedure is that up to this point we've been able to pack the plants together much of the time at a density of about 25 per square foot (250/m<sup>2</sup>), and they've been in the greenhouse for an optimum total of eleven weeks. During this brief period they have been bedded, planted, grafted, and established — all of this for an eventually large, woody, sensitive tree! Of course our efficiency is not always up to this standard; our plant densities fall off from time to time as we sort, cull, and box plants for readiness, but generally it is concentrated, efficient production.

Before proceeding to the field planting of the small grafted liners (our term for the plant at this stage) it is appropriate to describe the greenhouse conditions. Our choice for the greenhouse environment is not scientifically based. We simply

try to approximate the atmospheric conditions one would expect during an ideal coastal southern California April. Accordingly, we run a nighttime temperature of about 55°F (13°C) and a daytime temperature of about 72°F (22°C). We keep the relative humidity in excess of 55%. Our night temperatures are allowed to fall, principally, because low night temperatures aid in developing a suitable girth/height ratio in our plants. We believe this is important because it facilitates grafting, minimizes shading under crowded conditions and, we believe, affects the eventual girth/height ratio of the saleable tree. It is a ratio which we seek to maximize.

Transplanting of the grafted liners to field containers generally is done in the spring after the worst danger of frost has passed. For field containers we use open-ended flexible polyethylene sleeves of 10 mil material (.001 inches, or .025mm, thick), that measure 7" (18cm) in diameter by 17" (43cm) in height, when filled with field container soil. They are placed in rows that are two containers wide, the rows being on 38" centers, and then filled in place by hand with the aid of a special portable frame. We use open-bottomed containers for two reasons: first, they are cheap and simple to fill and, second, the intimate contact between container soil and the underlying earth assures efficient drainage of the container growing medium.

Field container soil is for us a compromise. To achieve efficient drainage and aeration we use 50%, by volume, of sawdust, 10% mushroom compost, and 20% sand. To achieve a certain degree of cohesiveness in our ball of soil, so that it may be transplanted efficiently, we include 20% clay-bearing loam soil. The clay component helps, too, with its ability to retain moisture.

Grafted liners are planted into the permanent field containers by slitting and stripping the plastic bags away from the young root systems (taking care to slice off any curled roots) and lowering the small balls into holes previously bored in the upper surfaces of the field containers. Success of transplanting is about 98%. The grafted liners needn't be large for this operation; 1/2" (150mm) of healthy scion growth is sufficient.

We have traced the small grafted liners' history now, from the fall of one year to the following spring, when they are transplanted to field containers. The trees will be ready for orchard planting the following late summer (10 months from seeding) or, more suitably, the following spring (15 months from seeding).

Field growing is conventional. The young plants are irrigated two or three times per week with a spaghetti-type drip

system under a constant feed program. They are staked and tied. Staking occurs as late as practicable since the later the staking, the higher the girth/height ratio that results. Mature, saleable trees are two feet (60cm) tall minimally, they average about three feet (90cm).

## DISEASE CONTROL

Avocado nursery diseases can be partitioned into two groups; 1) those whose presence we can't tolerate on saleable plants, and 2) those that threaten our production efficiency. The first group may not encumber our production efficiency but may raise havoc in a commercial orchard. The second group presents economic and marketing problems for the nursery by reducing yield and stunting the plants.

The first and most important disease that we cannot tolerate in a saleable tree is avocado rootrot, cause by *Phytophthora cinnamomii*. The fungus must be *absolutely* banned from our growing grounds. This is *extremely* important, as the fungus is readily communicable and kills mature orchard trees, often in a very short period of time. We, and many other nurseries, operate under a California State Certification program which stipulates many of the precautions we must take to avoid contamination of the trees. Among our precautions and, in addition to other conventional disinfecting and prevention procedures, are the following:

- A. Preplanting heat treatment of all seeds in a hot water bath for 30 minutes @ 120-122°F (49°-50°C).
- B. Fumigation of all soils, peat moss, and other soil amendments, @ 800lb/A (900Kg/Ha) or 3 lb./100ft<sup>3</sup> (48 g/m<sup>3</sup>).
- C. Dry copper sulfate (plus flocculent) step basins to be used by all entering foot traffic.
- D. Entrance water vats for wheel traffic, charged daily with formaldehyde.
- E. Weekly copper sulfate sprays on all greenhouse floor areas and bench legs, and on areas where outside traffic approaches our entrance areas.
- F. Sodium hypochlorite-bearing high pressure spray to sweep clean and disinfect the beds of all trucks before they pick up trees.
- G. Planting of susceptible indicator plants (*Persea indica*) among our regular outside field stock.

A second disease that we try to avoid spreading in nursery stock is sunblotch, a yellow and/or pinking streaking of stems and fruit in bearing trees. It is a systemic disease, thought to be viral in nature, which is transmitted through seeds and scionwood. To avoid it, we are more and more using Registered

sources of seeds and graftwood. These sources are the direct descendants of trees that have been indexed for detection of the disease under the supervision of the California Department of Agriculture and the University of California at Riverside. This disease is not nearly so important as avocado rootrot since it doesn't spread rapidly to otherwise clean fruit-bearing trees.

A minor parasite that we control is *Latania* scale. When passed on to orchards it tends to thrive on and girdle young tree trunks that are enclosed in poly-coated sun shields. We control the pest quite effectively with trimonthly field applications of malathion.

Our principal nursery pest (not of known major importance from the standpoint of spreading to orchard sites) is *Rhizoctonia* organisms. These destroy seeds and young seedlings and stunt root systems. They make liabilities of otherwise efficient operations in short order. We fight such potential infection intensely. In addition to other conventional disinfecting techniques we do the following:

- A. Treat all seeds, after heat treatment, serially, with sodium hypochlorite and a Benlate dip.
- B. Drench all bedding soil with Benlate and Terraclor.
- C. Treat all graftwood with Benlate.
- D. Drench all greenhouse planting media with Terraclor at time of planting, and all greenhouse media and grafted liners as a matter of course on a trimonthly schedule.
- E. Often in winter and spring, we drench the large field containers with Terraclor as the grafted liners are planted.

Our practices have functioned very well with regard to *Rhizoctonia*. Even in our susceptible fumigated soil we are troubled very little by it.

## EXPERIMENTAL WORK

As any commercial nurseryman knows, experimentation with new techniques is expensive and it usually fails, yet it is absolutely essential if the nurseryman wants to be among the industry's leaders. Also, it's one of the true joys in our work.

I will report a couple of items which may be new and useful. The first deals with seed storage and the second, a commercial method for clonal rooting of the difficult-to-root avocado rootstocks.

We tried for several years, with very poor and variable results, to store large quantities of avocado seeds under refrigerated conditions in polyethylene bags. The University of California could do it in small amounts, a few nurserymen we knew of could do it in small batches, but neither we nor any of our col-

leagues could dependably store large quantities of avocado seeds for many consecutive months. We noticed that entire bags of seeds died as if they were poisoned by their own respiration products. Therefore, we tried a form of periodic air exchanges. We inserted two stoppered tubes in the mouth of each bag, each extending from depths of the bag to the atmosphere. Monthly thereafter, we circulated atmospheric air through the bags with the aid of an air compressor, via the temporarily unstoppered tubing. In addition, we used thin plastic (1-1/2 mil) for bagging material. Since we began this practice, we have not experienced the death of any entire bags of seeds, and the viability rate has remained very high (90%+) for 6 months. We still don't know the limit of such storage, as 6 months is the longest period we've tried to date.

One of the most exciting projects we've worked on at Brokaw Nursery is the development of techniques to commercially produce clonal avocado rootstocks. This task became important some years ago when Dr. George Zentmyer, of the University of California at Riverside, discovered that certain clones, used as rootstocks, demonstrated significant resistance (or tolerance) to *Phytophthora cinnamomi*.

The most significant early work in the clonal rooting of avocados was done by Ted Frolich at UCLA (2). Frolich used the etiolation of potential rooting tissue as a powerful tool, new to avocado propagation. Basically, he grafted seedlings with bud-bearing stem tissue from a clone to be rooted, raised the grafted shoot to about a six inch height (15cm) in moist darkness, and enclosed the etiolated stem in a rooting medium, with the seedling still attached. After leaves had developed on the stem (now extending above the rooting medium) he severed the etiolated graft from the seedling and rooted the resultant cutting by conventional rooting procedures in vermiculite within a humidity chamber. After the rooted cutting had been established in a new container, he grafted it with a fruiting scion. Thusly he grew a grafted avocado tree of certain genetic identity in both rootstock and scion portions.

With this technique, Frolich was able to raise thousands of clonally stocked trees for Zentmyer's very important work on rootrot resistance. The commercial difficulty with the technique was that no commercial nurserymen could use it to produce large quantities of trees and, at the same time, make a profit. It was too slow, required too much space, and there were many failures.

Three years ago Brokaw Nursery hired Burton Silva, a recent graduate of Cal-Poly, Pomona, among other things, to administer a program to produce clonal avocado rootstocks. As a

result of Silva's work we were able to employ a method which is an extension of techniques previously worked out by Frolich, with an added twist.

The method departs from Frolich's at his cutting stage, at which he severs the new potential stock from the seedling. We have substituted a "weaning" process at this point to avoid shock to the plant and to utilize the vigor of the nurse seedling as long as possible. Our steps are the following:

1. Graft a seedling in the greenhouse with a bud-bearing stem scion of the clone to be rooted. (Frolich's procedure.)
2. Etiolate (grow in the dark) the rootstock stem to a height of 5-6" (13-15cm). (Also Frolich's procedure.)
3. Slip a wide metal collar, about 1/4" (64cm) in diameter, loosely over the etiolated stem to a point immediately above the graft union. (This metal band is called the *weaning girdle*.)
4. Extend the growing medium (now the rooting medium) nearly to the apex of the etiolated shoot.
5. Allow the grafted shoot (whose lower portion is etiolated) to develop to a graftable stage.
6. Graft a fruiting scion onto the clonal rootstock shoot.
7. Transplant the three stage plant in its entirety into a larger container for development to a maturity suitable for orchard planting. During the process the new rootstock is automatically weaned from its nurse seedling, which dies.

Rooting takes place in the above process sometime during stages 4-6, depending upon the girth of the plant. Silva's raw data suggests that over 90% of all etiolated stems have rooted after reaching a diameter of 7mm, one inch above the rooting medium.

The steps above are amenable to minor changes. Certain plastics may be substituted for the metal collar, but this is a detail only. We prefer metal because it girdles the stem very assuredly with less chance of the girdle's "callusing over" than with corresponding widths of plastic. We have never seen incomplete girdling where our metal bands were used.

In step three it is important not to rupture the cortex of the etiolated shoot. If the shoot is damaged in avocados it may callus to the point of bridging the girdle.

This procedure is currently protected by a Method Patent for avocados. It is now used commercially by Brokaw Nursery in the United States and (by special exclusive licensing agreement) by Getzler Nurseries in Israel. The method may be applied freely to difficult-to-root species other than avocado and



perhaps has some usefulness among them. It is similar to a method described by Hartmann and Kester (3), but the published technique does not dependably serve for avocados.

### FUTURE AVOCADO RESEARCH

I believe that in the future our research efforts will be directed toward the search for other avocado rootstock clones for a variety of specific purposes. We need rootstocks, for instance, with better rootrot resistance, rootstocks with dwarfing tendencies, and stocks that withstand salty soil and water, and for calcareous soil conditions. Techniques for economic, commercial, clonal rooting, are now in operation, and these will be surpassed in the future. We now need to find suitable stocks for special problem areas so that we can truly match special conditions with special plants — tailored from top to bottom.

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JIM POORBAUGH, Moderator: Do we have any questions for our speakers at this time?

BRIAN GAGE: It was stated that Monrovia Nursery brought quantities of peat moss into the grafting house. What was the peat moss used for? I assume it was for plunging containers.

DENNIS CONNOR: We are bringing in peat moss that has been wetted down and treated with captan into the grafting tents to supply the humidity for the grafts once they have been prepared and put in the tent.

ED SCHULTZ: Mr. Connor, do you use bottom heat on your juniper cuttings when you put them outdoors for rooting?

DENNIS CONNOR: Yes, we do. We are starting out at about 65°F bottom heat for most of the conifers that we produce. In about a 6-week period we are upping that to about 70° to 72°F. However, we have been doing a lot of experimenting in the past few years trying to get away from the use of bottom heat; this seems to be O.K. on certain kinds of conifers that even do better without bottom heat. For example, some of the "spreaders," like Bar Harbour, will root a little slower but just as well without bottom heat. It is a space problem for us at Monrovia with the amount of junipers we cut. We don't always have the bottom

heat space to put the plants in we would like to. So when we plan our production of junipers for the year, we try to arrange them so that the kinds that have to have bottom heat will go on the bottom heat beds, while the plants that can do without it will go on beds without bottom heat.

VOICE: What type of wood are you using to prepare conifer cuttings?

DENNIS CONNOR: Basically it is semi-hardwood. This is true for all the conifers we produce.

ARDA BERRYHILL: What is the rooting mixture you use?

DENNIS CONNOR: We use a 90% perlite and 10% peat, which is standard for our entire cutting operation. We use that mixture for just about everything that we propagate, with a few exceptions; for example, on azaleas we use a little more perlite of a finer grade. A few kinds of plants we propagate in sand. But basically the perlite-peat mix is used for everything we propagate.

HOWARD BROWN: A question for Mr. Fielding. What would you consider the best source of seed for cactus propagation?

MR. FIELDING: You can't find cactus seed listed in the usual seed catalog. It is a specialty, of course. There are cactus growers who make a business of doing nothing but growing plants and collecting the seed. The average type of seed will run \$2.50 per thousand, but some is \$1.00 per thousand. The growers in Fallbrook, California, raise a lot of cactus seed. Seed that is collected from Mexico to South America is harder to come by but there are people there who specialize in cactus seed. New Mexico Research collects seed from all over the world. They have a catalog and you can buy seed from them. You can also buy seed from Germany and England that is collected all over the world.

BILL NELSON: I would like to ask Mr. Brokaw about his experiences with dwarfed types of avocados, genetic or otherwise.

WILLIAM BROKAW: Dr. Hartmann, in his talk, mentioned the genetic dwarfs; we have these in avocado — several types which tend to be dwarfed. We are trying to find dwarfing rootstocks, but we haven't run into one yet. The problem is in recognizing them because what we are necessarily working with most of the time are seedlings which often look like they are dwarfed. They are dwarfed seedlings all right, but when we graft a fruiting scion on them they get big. We have a standing inquiry out to the industry to let us know if anyone finds a tree of a standard cultivar on good soil which is small and bears

well. We would sure like to force roots out from the rootstock shoots and duplicate the trees, thus obtaining a dwarfing rootstock for avocados.

BRIAN GAGE: Is there any research being done on inducing hardiness in avocado by the rootstock?

WILLIAM BROKAW: There is no evidence that rootstocks have anything to do with the cold hardiness of the fruiting scion of an avocado tree, I am sorry to say. We have some fairly hardy strains, such as Yama, which are suitable in the San Joaquin Valley of California. This one doesn't bear too well but does stand quite a lot of frost.

FRANK EVANS: Question for Mr. Brokaw. Does the Mexican avocado seed require treatment from the Department of Agriculture before you get it?

WILLIAM BROKAW: No, but as a matter of course, we are more and more using seeds which are from registered sources which has to do with the sun blotch that I mentioned; then as part of the certification program, we must be supervised in the heat treatment of the seeds before we put them into certified nursery locations.

LES CLAY: Question for Dr. Hartmann. I would like to know if he has any record of what work is going on in tissue culture in England.

HUDSON HARTMANN: There is considerable work on tissue culture being done in England. *Malus* (apple) has been propagated in England in this manner. I can send you a copy of the reprint describing this work if you are interested in it.

LES CLAY: My father was in England earlier this year and he came back all excited about two-year-old apple trees there with full crops of apples on them which he saw.

HUDSON HARTMANN: This is the "meadow culture" which has been developed at the Long Ashton Research Station near Bristol. They are planting the trees in an ultra-high density pattern and by the use of certain growth regulators and dwarfing rootstocks they bring the trees into bearing very early and are getting high yields. Whether this will develop into something commercial remains to be seen. But there is a lot of work in tissue culture going on in England. For example, Dr. Street in England was one of the leaders in tissue culture research. His book, *Plant Tissue and Cell Culture*, has just been published in its 2nd edition.

VOICE: Do you know of any avocado strain where the fruit can be left on the tree for about a year and a half before it is harvested.

WILLIAM BROKAW: No, but you could certainly get full year-round production by planting two cultivars and heavy two-year production with three cultivars. I would recommend planting three trees of different cultivars in one hole.

VOICE: Where does DNA recombination fit into tissue culture research in the future and in plant hybridization.

HUDSON HARTMANN: We are going to have a talk on Friday on this subject so if you can stay until then you will, perhaps, get all your questions answered. Cell and tissue culture research is a new field but DNA recombination is probably quite a ways down the road for plant propagators, although it is something coming up in the future. Genetic engineering, where you might develop your own plant, is a fascinating idea, but it is not in the immediate future for plant propagators.

VOICE: What is the strength of the Clorox solution that you use for avocado seeds; a second question is whether or not you condition the seed before dipping in Clorox.

WILLIAM BROKAW: We use 10% of the 3% household bleach. The answer to the conditioning is essentially — no. What we have is kind of a cold water dip, then we dip them into the Clorox solution. Shortly after that we give them a Cap-tan treatment.

VOICE: At what time, if ever, do you excise the original nurse seedling in your nurse root grafts?

WILLIAM BROKAW: Never, it is automatically aborted. With the nurse root graft procedure we use now the root is automatically cut off and we have never found a failure.

## **POTENTIAL APPLICATION OF PROTOPLASTS FOR FUTURE PLANT IMPROVEMENT**

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### **INTRODUCTION**

Traditional plant breeding methods for commercial improvement of plants are restricted to hybridizing plants that are closely related. With few exceptions, hybrids combining desired qualities derived from both parents can only be made between different species of plants. These  $F_1$  hybrids are usually self-sterile and require doubling of the chromosomes before they can be used for further breeding purposes. Intergeneric hybrids are very rare. In recent times, some notable discoveries have come out of fundamental research on plant tissue cultures.