PRESIDENT BRUCE USREY: To start the meeting I thought we could have a kind of self-introduction. So if each of you would stand up, give your name, where you are from, and your speciality, the thing you really enjoy doing, then we will get the program going. This will help us get acquainted right from the start.

Thank you. Now Bruce Briggs, our first Moderator, will start the program.

MODERATOR BRUCE BRIGGS: We will now hear from the first speaker, Steve Wong, introducing our first session on "Tissue Culture after the Test Tube":

## DIRECT ROOTING OF TISSUE-CULTURED RHODODENDRONS INTO AN ARTIFICIAL SOIL MIX

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Abstract. Direct rooting of tissue-cultured rhododendrons into an artificial soil mix was highly successful. By by-passing the standard test tube pre-rooting stage in the laboratory, production costs are markedly reduced. If a suitable environment is maintained in the propagation tent, root initiation will occur within 3 to 4 weeks of planting.

It has been accepted by most propagators that tissue culture propagules must be placed into a pre-rooting stage to condition them for transfer to a soil medium. In the prerooting stage the propagules are placed in a charcoal medium in the test tube for about 4 to 6 weeks to allow for an increase in size, uniformity, and hardiness. This makes for easier planting, but in our experience, only slightly higher survival rates. Motivation for bypassing this pre-rooting was to reduce costs by eliminating this one additional operation and the attendant use of expensive test tubes and media, aseptic techniques, and laboratory equipment and space. In our initial attempts at rooting directly into an artificial soil mix, the planted flats were in styrofoam boxes with a glass top (1). Humidity was maintained with manual misting throughout the day. Results were promising so we decided to try this on a larger scale in a propagation tent. Results have been outstanding, convincing us that direct rooting in an artificial soil mix is both physically possible and economically feasible.

## MATERIALS AND METHODS

The process of establishing the unrooted tissue culture propagules directly into an artificial soil mix involves four operational stages:

- 1. cutting up the propagules
- 2. planting in the artificial soil mix
- 3. rooting and growth in the propagation tent
- 4. potting
- 1. Cutting up the propagules. The clumps of shoots are removed from the test tubes with forceps, washed, and then cut for planting. Callused growth and undersized, deformed, and watersoaked propagules are discarded. Well-formed shoots 75 mm and up are used for planting. On the average, 15 to 30 plantable shoots are obtained per test tube. With this method, the cutting does not need to be done aseptically in the laboratory.
- 2. Planting in the artificial soil mix. This is extremely meticulous and visually straining work and workers should be hired with this in mind. The inside of our planting flat measures  $47 \text{ cm} \times 30 \text{ cm} \times 4 \text{ cm}$ , with plantlets at a high density spacing of  $2.0 \text{ cm} \times 2.5 \text{ cm}$ . It takes approximately 45 minutes to plant a flat of 285 propagules. A peat/perlite/sawdust mix is used, sieved to accommodate the small propagules. After planting, the propagules are given a hand mist of 20:20:20 fertilizer solution plus Benlate, then put into the propagation tent.
- 3. Rooting and growth in the propagation tent. It is imperative that there be strict control over the temperature, humidity, and light levels in the propagation tent. We arrived at the style of our present propagation tent through a series of preliminary trials. The tent consists of a plastic sheet supported over a 2.4 m  $\times$  19 m heated concrete bench, which accommodates 277 flats, or 79,000 propagules. The air temperature is maintained at 21 to 24°C during the day and 18°C at night. The soil temperature is kept at 24 to 27°C. Ventilation is with an exhaust fan and cooling pad arrangement.

We use high pressure sodium lamps as supplemental light to increase the daylength to 16 hours and the intensity to 5400 lux. To prevent an excessive heat load in the summer we blocked out the daylight with opaque plastic and have gone exclusively to the artificial lights. We have found that the moderate expense of operating these lights is more than offset by the increased rooting and growth.

Because these propagules are without roots and a cuticle layer, the humidity within the tent must be kept high to

prevent desiccation. This is achieved through frequent misting and use of a cooling pad. Our misting is automated and set for a short duration of 2 seconds every half hour during the day; however, if the weather is hot and the exhaust fan is on, the misting is increased to as much as every 8 minutes.

Under these conditions, some of the easier rooting cultivars, as 'The Hon. Jean Marie de Montague', 'Daphnoides' and 'Vulcan', will initiate roots after only 3 weeks. Cultivars like 'Britannia', 'Scintillation' and 'Cynthia', will take approximately 4 weeks. A flat of 285 'Daphnoides' was checked 35 days after planting and 64 percent of them had rooted. Almost all cultivars tested are ready for transplanting within 3 months of planting, with vigorous growers like 'Daphnoides' or 'Jean Marie de Montague' ready within 2 to 2½ months (Table 1). At transplanting time, the roots are well branched and in most cases over 1.5 cm long.

We have noticed that rooting will only occur if the propagule is in a succulent, juvenile stage. If there is environmental stress the tissue quickly hardens and root formation becomes extremely difficult, if not impossible.

A hormone powder was tested but did not seem to cause earlier root initiation although it did seem to cause a greater proliferation of roots. Further experimentation is needed to justify the additional labour of dipping the base of the propagules into the hormone powder.

Table 1. Rooting percentage of propagules at transplanting.1

| Cultivar                        | Days from planting to transplanting | Percent Rooted |
|---------------------------------|-------------------------------------|----------------|
| The Hon. Jean Marie de Montague | 56                                  | 90             |
| Daphnoides                      | 62                                  | 89             |
| Britannia                       | 62                                  | 81             |
| Les Clay                        | 75                                  | 73             |
| Scintillation                   | 75                                  | 79             |
| Vulcan                          | 76                                  | 66             |
| Golden Witt                     | 77                                  | 89             |
| Cotton Candy                    | 83                                  | 80             |
| Rainbow                         | 84                                  | 73             |

<sup>&</sup>lt;sup>1</sup> 285 propagules of each cultivar planted in May and June, 1981.

4. Potting. The rooted plants are put into a peat/perlite/sawdust mix in 2¼" pots. They are kept under mist for about 1 month before moving into shade houses. Tissue-cultured plants grow much more vigorously and are better branched than cutting-propagated plants. They are ready for sale approximately 3 months after potting.

#### RESULTS AND DISCUSSION

From our experience so far, we feel that it is feasible to bypass the pre-rooting stage and have the propagules go directly into an artificial soil mix. If a suitable environment is maintained there is a high percentage of root initiation within 3 to 4 weeks. To include the pre-rooting test tube stage would mean handling the plantlets an additional time in the laboratory, increasing production costs.

## LITERATURE CITED

1 Anderson, W.C. 1978. Rooting of tissue-cultured rhododendrons. Proc. Inter. Plant Prop. Soc. 28:135-139.

# TRANSPLANTATION AND POST-TRANSPLANTATION OF MICROPROPAGATED TREE-FRUIT ROOTSTOCKS

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**Abstract.** The principal factors that have affected success at transplantation of micropropagated tree fruit rootstocks are presented. The factors discussed include pre-transplantation conditions (laboratory culture) and the conditions at transplantation and field-planting; examples are taken from the Kelowna Nurseries production.

Abbreviations used in the text:

NAA naphthalenacetic acid
IBA indolebutyric acid
IAA indoleacetic acid
BA benzyladenine
MS Murashige and Skoog

Murashige and Skoog nutrient formulation (8)

The use of micropropagation as a propagation tool is often viewed with skepticism. More specifically, it is the rooting and transplanting phases which need credibility. These stages present challenges at least equal to the initiation of cultures, and are more awesome because they mark the end of artificial control and a return to more traditional practices.

Recent publications concerning transplantation (2,3,6) and the factors influencing rooting in tree-fruit culture (5,9,10,11) give indications of diverse approaches.

Our approach is based upon survival and growth after transplation which relies on in vitro cultural history coupled with horticultural practices as follows.