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The Role of Micropropagation in the Incidence of Tissue Proliferation in *Rhododendron* 'Montego'

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Tissue proliferation (TP) is a condition primarily seen in certain micropropagated rhododendrons. Cultivar Montego was studied because plants grown in containers and in vitro cultures show distinctive morphological and physiological phenotypes which identify plants or cultures which are prone to TP.

Our goal in this study was to test the hypothesis that adventitious events during micropropagation are involved in the induction of TP-like culture characteristics and to test if the cytokinin 2iP plays a role in this induction.

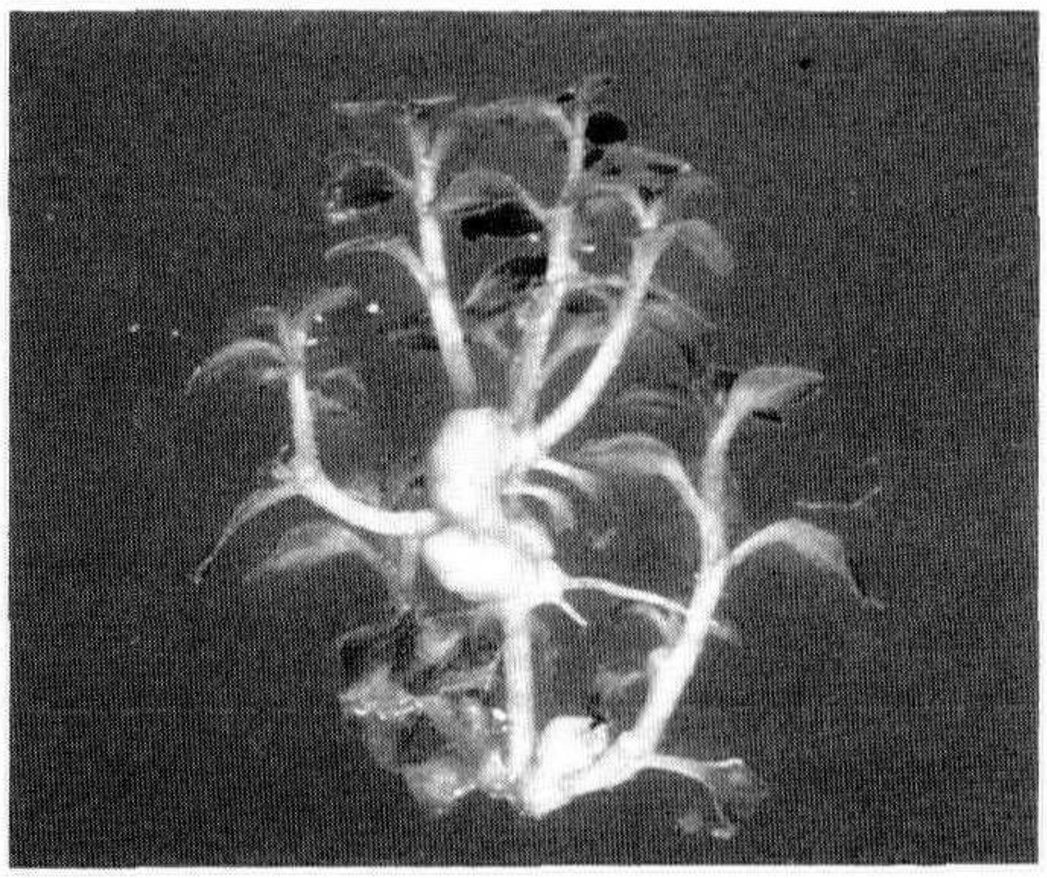

Figure 1. Swellings produced on shoots of a TP+ in vitro culture of *Rhododendron* 'Montego' on basal Woody Plant Medium.

Comparisons were made between 'Montego' with TP (TP+) and 'Montego' control plants that had no TP (TP-) in their propagation history. This was done to avoid the possibility plants or tissues derived from plants with TP may retain the potential to express TP even if TP was not apparent. TP- stock plants were obtained by rooting cuttings from the original 'Montego' plant grown from seed. Cuttings were supplied by Dr. David Leach of the Holden Arboretum. TP+ 'Montego' plants or tissues originated from plants with TP.

Three experiments were conducted. Experiment 1 compared in vitro culture initiation and maintenance char-

Figure 2. Typical TP+ in vitro culture of *Rhododendron* 'Montego' growing on basal Woody Plant Medium.

Figure 3. Typical TP- in vitro culture of *Rhododendron* 'Montego' growing on Woody Plant Medium containing 10 mM 2iP.

acteristics of TP- and TP+ explants; Experiment 2 tested the effectiveness of 2iP in inducing TP-like characteristics in TP- cultures; and Experiment 3 studied the role of adventitious events in TP- tissues in generating TP+ culture and plant characteristics.

Cultures were initiated by transfer of explants from TP+ and TP- stock plants onto Woody Plant Medium (WPM) supplemented with 10 mM 2iP. Shoot tips of TP+ and TP- 'Montego' behaved differently after the culture initiation period. TP+ cultures miniaturized and multiplied 4 months after initiation (AI); TP- cultures required 10 months to reach the same stage. Dramatic changes occur in TP+ at 6 months when excess shoot branching with little shoot elongation and very rapid multiplication began. This is accompanied by development of TP-like swellings at nodes (Fig. 1). By 10 months TP+ cultures become so compressed due to reduction in shoot extension they were overwhelmed by adventitious growth. In order to encourage shoot extension, we then transferred shoot clusters off initiation medium onto basal WPM which contains no hormone. Growth on basal WPM was required to maintain TP+ shoot cultures (Fig. 2). Repeated subculturing shows these cultures can rapidly multiply in the absence of 2iP. This culture behavior is typical of TP+.

TP- cultures maintained on WPM with 10 mM 2iP begin slow, steady multiplication 14 months AI. Only nonadventitious shoot tips and stem segments were transferred. The rate of multiplication has not increased 28 months AI, and 2iP is required for multiplication. Shoots elongate well, but multiply slowly (Fig. 3). This culture behavior is typical of TP- cultures.

In Experiment 2 significantly more adventitious meristem was produced at the base of TP- shoots grown on WPM containing 50 mM 2iP than on WPM containing 10 mM 2iP. The increase in adventitious meristem was correlated with an increase in TP+ culture behavior.

The role of adventitious events in generating TP-like characteristics was further investigated in Experiment 3 where shoot organogenesis from TP- leaves was used to generate adventitious growth which was classified by phenotype as putative TP+ or putative TP-. Six of 41 leaves (15%) of the leaves produced cultures with TP+

culture behavior (putative TP+). The presence of intermediate cultural phenotypes suggests that partial conversion of TP- to TP+ may occur during adventitious events. Plants grown from putative TP+ and putative TP- cultures produced significantly different leaf and shoot morphology. This morphology was statistically identical to control plants derived from TP+ and TP- stock plants and cultures initiated from their shoots.

Many growers are concerned that micropropagation cannot produce plants free of TP. Our work shows that phenotypically normal rhododendron can be produced via micropropagation. We have not observed TP+ tissues arising from axillary multiplication from nonadventitious shoot tips. Stable, "normal" cultures can be maintained by: (1) vigilant removal of basal callus and adventitious meristem masses, (2) use of low cytokinin levels to maintain slow-to-moderate multiplication rates, and (3) production of shoots through axillary multiplication from nonadventitious shoot tips.

Utilizing Band Pots For Herbaceous Plant Production

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Purpose. Band-pot technology is not new to plant propagators. Indeed, this type of container, originally known as milk carton blanks or pots, has been in use for many years with several variations of different types of materials. This presentation shows the utilization of this container to grow herbaceous plants and the subsequent uses for this band-pot-grown product.

Why Use Band pots. I looked at using band pots after considering several factors.

1) Presently, most of the mail order nurseries sell their perennials, bare root. Customers receive their plant material via mail or UPS. They then either pot it up or plant it into their gardens. People in most cases pay full retail prices for a bare-root plant.
2) In general, most perennial crops are 8 to 12 weeks in production duration after transplanting seedlings or rooted cuttings. It is possible to grow plants in band pots and bring them to marketability several times during 1 year or season.
3) The reutilization of containers can effect lower production costs. This of course means that you could gain a better profit margin, if all aspects of production remained fixed or the same.
4) You could in effect plant a "smaller" container on a project, yet still get "1 gallon" results after reasonable period of time.

What I am trying to do with my company is grow a reasonably priced plant for a retail client, yet keep my costs as low as possible. Considering that mail order companies sell bare-root materials for full retail prices, or in some cases better than, I tried to incorporate this concept into a price and product formulation for local clientele. Since my present production location is limited in size, I had to get the most material grown in a limited space—about 1 acre. Field growing plants and then bare root storage was out of the question. So I decided to begin growing seedlings or divisions, depending on size, and cuttings in a smaller, yet marketable container. The bottomless band pot seemed to fit the bill. They come in a range of sizes, allow