What's New In The Biology Of Adventitious Root Formation

Fred T. Davies, Jr.
Texas A&M University, Dept Horticultural Sciences, College Station, Texas 77843-2133

INTRODUCTION

The First International Symposium on the Biology of Adventitious Root Formation was held in Dallas, Texas, 18 April through 22 April 1993, with over 140 participants, representing some 25 different countries. It was a unique opportunity for researchers from industry, government, and many different academic disciplines to get together and focus on the basic and applied aspects of adventitious root formation. The Southern and Western Regions of IPPS helped sponsor the symposium.

No participant had a magic bullet or quick solution to rooting problems, and more questions were raised than answered. Inability to agree on a model system was probably a healthy sign and points to the complexity of adventitious root formation.

COMMERCIAL IMPORTANCE OF ADVENTITIOUS ROOT FORMATION

Vegetative or clonal reproduction is the most important propagation method of many horticultural crops. United States of America horticultural industries comprise one of the largest combined commodity crops in agriculture. In fact, the USDA reported that in 1991 export value of horticulture crops exceeded all other agricultural commodities, including forestry and agronomic crops (USDA, 1991). The 1991 United States of America wholesale value of ornamental crops (nursery crops, greenhouse, floriculture, foliage and bedding plants) was $8.9 billion, while total consumer expenditure for ornamental crops was $40 billion.

ADVENTITIOUS ROOT FORMATION IN HORTICULTURAL CROPS

Over 70% of the propagation systems used in the ornamental horticulture industries depend on successful rooting of cuttings. The large international floriculture industries such as poinsettias, carnation, and chrysanthemums rely exclusively on asexual propagation by cuttings.

In southern and western U.S. ornamental nurseries, seed propagation typically accounts for less than 10%; division from 5% to 15%, and graftage, spores and tissue culture liners less than 1% of the propagation systems utilized, compared to 70% to 90% propagation by cuttings. Ornamental shrubs are usually asexually propagated by cuttings, whereas shade trees are propagated by seeds due to difficulty in rooting. Easy-to-root plant materials are much more widely propagated as tissue-culture-produced liners than are difficult-to-root species. Even though tissue culture can enhance a difficult-to-root woody species' ability to be cloned, poor adventitious root formation often limits their commercial production through tissue culture systems.

LIMITATIONS OF ASEXUAL REPRODUCTION

Adventitious root formation is a prerequisite to successful clonal regeneration of propagules, with possible exceptions of apomictic seed, and graftage and budding systems on seedling rootstocks. Poor rooting continues to be a serious commercial limitation in the asexual propagation of many woody horticultural crops. Propagation systems commercially utilized to improve rooting success such as mounding, stooling, layering, division, separation, graftage, budding, tissue culture, manipulation of stock plants through etiolation and banding are costly and labor-intensive.

Labor costs contribute to 30% to 65% of ornamental crop production expenditures and more than 80% of propagation costs, which gives considerable financial incentive to streamlining techniques and improving rooting. Direct sticking cuttings into small liner pots as opposed to sticking into conventional flats uses personnel and materials more efficiently. The additional production step of transplanting rooted cuttings is eliminated, and the potential transplant shock due to a disturbed root system is avoided. However, hard-to-root species cannot be stuck directly into pots since rooting must be 80% or better to justify the additional space required. The inability to induce adventitious root formation seriously limits our ability to propagate many potentially valuable horticultural crops, particularly mature woody species.

GENETIC AND BIOCHEMICAL IMPLICATIONS TO ROOTING

Auxin is frequently not the limiting factor to successful rooting. So will magic chemical formulations for industry to use be available in the near future? Probably not. So we need to know what makes a cutting difficult to root. We know that many woody plant species are much more difficult to root when they become physiologically mature. Ability to form new cells, to respond to certain chemical stimuli, and to develop root initials are the most critical parts of the rooting process. A cell's ability to form new cells or remain meristematic, even while other cells are becoming specialized, or differentiated, may be determined during a stage of development when rooting would not ordinarily occur. The later development of root primordia and their elongation are rarely a limiting factor in rooting.

So what controls cells' meristematic ability? Much of the rooting research to date has relied on the chemical events and responses <u>after</u> genes have triggered the production of proteins or enzymes. Research needs to concentrate on the genetic processes controlling the production of proteins, some of which serve as important enzymes in the initiation and development of adventitious roots. Molecular studies at the DNA transcriptional and translational level have the greatest potential for revealing the control of rooting. Researchers in Wes Hackett's laboratory at the University of Minnesota have identified some biochemical markers for rooting such as polyamines, peroxidases, and flavenoids with the juvenile and mature forms of English ivy.

To date no current gene marker is clearly defined for rooting. There are phenylpropenoid genes (PAL, CHS, DFR), cell wall protein genes, and genes regulating cell division, but researchers do not agree that these genes are clearly defined for rooting.

Spano et al. in France worked with leaf explants of hairy root tobacco that were genetically transformed with the bacterium *Agrobacterium rhizogenes*. They were able to isolate different genes in the agrobacterium. They demonstrated that spontaneous rooting of the transformed tobacco plants was not due to auxin-producing genes or a substantially altered balance of endogenous hormones, but rather to those genes that increased the sensitivity of the tobacco to the presence of auxin. This has important implications in difficult-to-root woody species that have lost their rooting ability and will not respond to applications of auxin. The ramifications of this are that in the future, genes that enhance tissue sensitivity to auxin could be inserted into genetically transformed woody plant species.

ANTISENSE TECHNOLOGY AND ROOTING

The idea here is to switch genes on and off. When a plant undergoes changes from juvenile, to transition, to mature phase, its genome genes do not change. However, there are specific genes that are turned on or off change during the maturation process. These epigenetic, or nonpermanent genetic changes can negatively influence rooting since certain enzymes may or may not be produced. Antisense technology is currently being used in fruit ripening and in flower pigmentation where changes in flower color can be genetically induced. If an enzyme negatively affected rooting then an antisense DNA or RNA could be used to turn off the gene that produced the enzyme. What genes or gene groups affect rooting has yet to be determined.

POTENTIAL FOR EMERGING NEW PROPAGATION TECHNOLOGIES

Biotechnology includes whole-plant engineering using traditional plant breeding and selection, cellular engineering with cell culture and cell fusion and molecular engineering at the genetic level. Development of somatic embryos, called embryoids, from vegetative rather than reproductive cells holds promise for synthetic seed technology. A somatic embryo encased in a synthetic seed can be used in fluid drilling systems. Clonally regenerated plants are being propagated as seeds that will produce offspring identical to the parent. The bedding plant industry is highly interested in this technology. Calgene produced somatic embryos of celery, which can be fluid drilled into the soil, are being commercially used in Mexico. The forestry industry is very much interested in this process because it circumvents the maturity phase of the plant, when rooting is reduced, and the plant material can be handled as seedling transplants even though they are clones.

STOCK PLANT MANIPULATION

So until this "new" technology is available, propagators can use more conventional techniques. Brian Howard from the East Malling Horticultural Crops Research Center presented results of some excellent applied research on stock plant and stock block manipulations, such as hedging and pruning systems, for rejuvenating plants and enhancing rooting.

CONCLUSION

Successful rooting of species that are currently uneconomical to root by cuttings will give the industry new plant products and markets. Using biotechnology to manipulate genes to improve tissue sensitivity to auxin, as well as to improve tissue culture systems may be the way these species can become important in the ornamental industry. Successful rooting and acclimation of tissue-culture produced plants will need to be improved if biotechnology is to be commercially incorporated into the propagation and production of ornamental horticulture crops.