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Somatic Embryogenesis in Forestry: A Practical Approach to Cloning the Best Trees

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Preface: Tress as well as humans have two basic cell types based on genetic content: somatic cells and gametic or reproductive cells. Somatic cells, such as skin cells or the sapwood cells in a tree, have at least twice (2n) the base set of chromosomes. The reproductive cells (gametic cells) have a single (n) set of chromosomes.

World population growth has placed tremendous pressure on agriculture, both on agronomic and tree crops. These growing populations need to be fed, clothed and housed. For efficient plant production, large acreages that are dedicated to cultivation of single species are virtual invitations to their parasites and pests. Changing global climates, whether initiated naturally or by intervention of man, alter the agricultural potential of vast areas and entire countries.

Not only must we contrive methods for rapid, large-scale propagation of needed crops, we must also anticipate problems such as insect pest epidemics, demographic changes, and progressive droughts.

Obviously, selection of the best plants (identified by such as healthiest, fastest-growing; disease resistant, or other favorable characteristics) for scale-up is a good idea. These may scale-up by cloning and then be naturally or **artificially** bred among others of that species that were selected for complimentary positive **features**. For example, one clone that was developed for its **dis-**ease resistance advantages may be cross-pollinated with another clone developed to exploit its drought-tolerance.

Methods have been developed for cloning hundreds of commercially useful plant species. In these Ways, millions of individual plants can be **developed** rapidly in small areas on laboratory table-tops or in greenhouses. **Once grown to a stage and size that will survive outplanting** to the field, they may be used direcdy to produce a **crop** or may be used in breeding programs with other clones to combine the genetic advantages of each.

There are several methods that have been used successfully to clone trees. These include rooting of **cut-**

tings (stimulating roots to develop on a cutting), **organogenesis** (initiating many shoots from a single piece of plant tissue, then roots on the growing shoots), and embryogenesis (using a plant tissue from which to initiate many entire embryos, which have both pre-formed root and shoot embryonic structures, then maturing the embryos to fully formed baby trees). Generally, the last method is most promising over the other methods and for several reasons.

For rooting of cuttings, the source tree must be adequately juvenile to contain tissues that will respond to a rooting stimulus (**generally** a synthetic plant hormone). In trees this juvenility may be lost within the first few years of growth before we can identify the best individual trees in a stand from which to take cuttings. Rooting of cuttings is an unreliable process; the cuttings from a tree often do not root at all. **Organogenesis** and embryogenesis are tissue culture processes undertaken in the laboratory. Organogenesis is a **multi-step**, time-consuming, and labor-intensive process requiring the successive initiation and growth of roots and shoots from the same tissue source. Each initiation step and all **plantlet** growth between and after requires a different nutritional regimen. These many steps require many man-hours of labor. For these reasons, it is frequently not commercially competitive with the use of seedlings although the latter does not provide a clonal population.

Embryogenesis holds the greatest promise yet apparent to workers in the plant sciences. Here, a portion of plant tissue—often the entire young embryo (zygote) found within a seed—is placed in a nutritive liquid or atop a gelled preparation of the same or similar liquid. The cells in that zygote continue to grow, producing a larger mass of cells in which most or all have undergone reverse development, back to the cell form from which they arose in the fertilized egg cell. This process of cell multiplication to a mass of many embryos is called embryogenesis. Thus, we end up with a very large number of cells that are capable of developing into an entire plant.

Because they arose from the body [**'soma'** = 'body' (Gr.)] of that zygotic embryo, the process of cell multiplication to a mass of many embryos is called "somatic **embryogenesis**." Because all organs of the plant are present

from each embryo's almost-microscopic first appearance, that embryo may be developed to the whole-tree in a more direct and less costly manner.

There are several other and practical advantages to the use of somatic embryogenesis (SEM) as a means to manufacture or develop plant clones. SEM produces a virtually unlimited number of embryos as long as the culture is grown and maintained in a healthy manner in the laboratory. Any one culture may be subdivided to smaller portions and inoculated in fresh nutritive medium for continued, unlimited enlargement of the cell mass.

If one wishes to carry out experiments on the SEM, it is easy to extract a small or large portion of the culture for other studies while maintaining the original culture as it was and continuing to grow it as before. Somatic embryos may be encapsulated in a gelatinous material then dried and stored for later use as seeds. These artificial seeds could be maintained for many months to years under appropriate environmental conditions, such as cold and darkness for later sowing in appropriate sites. Pesticides may be incorporated within the encapsulating gel to suppress competing vegetation when the artificial seed is later sown. These seeds may be outplanted, maintained in storage, or returned to conditions, which once again support **embryogenesis** in order to develop another population of somatic embryos. Theoretically, all these processes may be continued indefinitely.

Another and extremely promising application of somatic embryogenesis, is the potential for genetic engineering of individual cells within the SEM and then selecting and maturing the engineered cell or a cluster of cells grown from that first engineered event to **fully-formed** trees for outplanting in the field. We may engineer using genes for disease resistance or faster growth, drought tolerance, or resistance to harmful chemicals such as those herbicides used to suppress competing vegetation in a stand of trees. Genes are being newly identified in many plants; some of these genes show great promise for expression in commercially important crops.

Somatic embryogenesis may prove to be a widely applicable **process** for crop improvement in agriculture and forestry 🌲