

**THE STATUS OF THE ART AND SCIENCE
OF ROOT INITIATION**

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Since man recognized the needs and benefits of reproducing plants asexually and, more specifically, propagating them by means of cuttings there have been many significant advances which have made the technique more efficient and have extended the range of plants which may be propagated by cuttings. I would like to review the techniques and advances, pointing out some of the major contributions, showing the blend between art and science, and finally demonstrating that there still remain areas in which answers are still wanting and opportunities for research abound.

Early propagators quickly realized that one of the major problems which must be solved when propagating plants by cuttings is to control water loss. The bell jar provided one solution. It provided a restricted volume of air which could become quickly saturated with moisture, either from evaporation from the media, or from water vapor transpired by the cuttings. When the water vapor in the air surrounding the cuttings reached the concentration of water vapor within the leaves an equilibrium was established and the net loss of water from the cuttings was essentially eliminated. The bell jar, however, was inefficient because of its small size and the fact that it had to be watched carefully to ensure that it was not exposed to direct sunlight. Not only did the small volume of air allow a rapid accumulation of moisture, it also acted as a heat trap and, if exposed to direct sunlight, would accumulate sufficient heat so as to severely damage the cuttings.

Grafting cases, or Wardian cases, were developed which facilitated handling larger numbers of cuttings and also increased the efficiency of operation in terms of shading and ventilation. This system was continuously used for many years with only slight modifications.

A specialized system developed for the propagation of rhododendrons was developed by Guy Nearing. The Nearing frame was essentially a cold frame with a fixed reflector. The reflector was designed to be open to the north and the frame was only exposed to reflected light. In this way the rhododendron cuttings would receive sufficient light by reflection and not be exposed to direct sunlight which would cause an undesirable buildup of heat within the frame.

The introduction of plastic film, or polyethylene, provided opportunities for new advances in efficiency and flexibility. The plastic

could be used to replace glass, which reduced the weight of the sash, and thereby facilitated handling. In many cases the conventional grafting case was substituted by a polyethylene tent, sometimes referred to as the vapor-proof case. Cuttings could be placed in the polyethylene tents, watered well and covered with the polyethylene, and essentially be left undisturbed until rooting had been completed. Up to this point, however, although there were changes in the mechanics of controlling water loss, the basic principle remained unchanged. That is—water loss was controlled by building up the humidity surrounding the cuttings.

In the early 1950's a new technique was introduced called, "mist propagation." Although some propagators had used a similar technique many years earlier by mechanically syringing the cuttings every few hours, mist propagation brought a degree of mechanization that was unique to the propagation operation. It also added a new parameter in the control of water loss from cuttings. That is, rather than just depending upon increasing the humidity around cuttings, the leaf temperature was decreased thereby reducing the tendency for water vapor to be lost from the leaves. This new dimension in moisture control also allowed propagation in higher light intensities and did not require a restricted air space. As a result, a wider range of plant materials could be propagated by cuttings, particularly when very soft cuttings were used; that is, cuttings from the new growth of plants. It had not been possible to use these cuttings previously because the soft nature of the tissues, coupled with a low initial reserve of carbohydrates, resulted in their quick decline and decay in the grafting case. But the new environment of mist with its cooler temperatures and higher light intensity facilitated both rooting and photosynthesis. Since the cutting tissues are highly active in the early spring flushes of growth, rooting took place in many cuttings formerly considered impossible to root. Mist propagation truly represented a major contribution to the vegetative propagation of plants. Since the introduction of mist propagation there have been changes and refinements. Initially mist was applied continuously and it was quickly found that an intermittent form of mist was superior. Intermittent mist reduced the leaching and water-logging problems that were associated with constant mist. Also intermittent mist did not reduce the medium temperature below optimum levels. Since the application of any water to cuttings will lead to some leaching, the concept of adding some nutrients to mist was introduced. Although in some cases nutrient mist has accelerated rooting, the greatest benefits appear to be realized in the subsequent growth of the cuttings once they are rooted. In addition to applying nutrients to the mist some researchers have incorporated slow-release fertilizers in the media. Materials, such as Osmocote and MagAmp, have had some beneficial effects on root initiation but again the greatest benefit is realized once the cuttings are rooted.

During the time propagators were concerning themselves with different ways of controlling water loss from cuttings another highly significant discovery was made. That was that plants contain growth regulators or hormones which can regulate or accelerate the process of root initiation. The isolation and identification of indoleacetic acid as the naturally occurring plant hormone led to the development of many synthetic growth regulators which were tested for their root promoting ability. Materials such as indolebutyric acid and naphthaleneacetic acid are now used routinely to facilitate the rooting of cuttings. Once again the range of plants which could be propagated by cuttings was enlarged and the efficiency of the technique was greatly improved. Again there have been some refinements and adjustments in the use of the plant growth regulators. They may be applied in talcum powder or in liquid, either as a concentrated dip or as a dilute soak. In most cases the concentrated dip provides efficiency of application and generally the largest response, since the growth regulator is already in solution and more rapidly penetrates the cuttings' tissues. Additives such as DMSO have been tried to facilitate the uptake and movement of the root promoting substances in the cuttings. Other materials, such as catechol, which appears to protect the plant growth regulators within the tissues of the cuttings, have been used. More recently a new group of plant regulators, commonly used for retarding the growth of plants, have been shown to have the ability to stimulate root initiation in some cuttings. The growth retardant, B-9, applied at a rate of 1000 to 5000 ppm has stimulated root initiation in chrysanthemum cuttings.

Although the introduction of mist propagation and the use of growth regulators have increased rooting efficiency and have extended the range of plant materials which may be propagated by cuttings, there nevertheless remains a large number of woody plants which are still considered to be nearly impossible to root.

Studies in the physiology of root initiation have been conducted to attempt to find the reasons why some cuttings are very difficult to root. It is clear that substances essential for root initiation are synthesized in the buds and leaves of cuttings and are translocated to the base of the cuttings through the phloem. The effectiveness of leaves and phloem transport in root initiation can be demonstrated by comparing the rooting of leafy and defoliated cuttings and cuttings that have been girdled compared to non-girdled cuttings. Leaf removal and girdling drastically reduce the rooting potential of a cutting. The substances which are translocated from the top of the cutting to the base include the naturally occurring auxin, indoleacetic acid, sugars, nitrogenous substances and a group of compounds which have been referred to as the "rooting cofactors." The cofactors are substances which react synergistically with indoleacetic acid to accelerate the rooting process. It has been suggested that the variation in

rooting ability of cuttings is an expression of how many and how much of the rooting cofactors are present. An easy-to-root cutting would have a good supply of auxin, carbohydrates and nitrogenous materials and rooting cofactors. Difficult-to-root cuttings may lack one or more of the essential components. In addition to the mobile components regulating root initiation, there appears to be a nonmobile component. It has been suggested that the nonmobile component may be a protein such as an enzyme. Research in Bouillenne's laboratory in Belgium indicates that the polyphenol oxidase enzyme may be involved in root initiation. The enzyme can be found present in those cells which are about to divide and differentiate into a root initial. We have found that the polyphenol oxidase isozymes appear in the hypocotyl of mung bean cuttings prior to root initiation and their development is accelerated when indoleacetic acid and catechol are supplied in the incubation medium.

In addition to the internal physiology of cuttings during root initiation the external effects of the environment have also been considered. The effect of light, both in terms of quantity and quality, have been investigated. It is interesting that light can influence rooting in strikingly different ways. For example, light is essential for photosynthesis and, in the case of softwood cuttings, increased rooting can be realized by increasing the available light. However, in the area of root initiation at the base of the cuttings, light can actually be inhibitory. In fact, if the tissues in which root initials are to be formed can be grown in the absence of light, that is etiolated, the rooting potential of the tissues is greatly increased. The etiolated tissues contain more free sugar, less starch, and essentially no lignification. The thin-walled cells appear to be easily induced to rapid cell division and differentiation into root primordia. As a general rule, long days—that is, 16 hours of light or more—favor root initiation and root development, while short days—8 hours of light or less—reduce rooting potential. The carbon dioxide content of the air surrounding the cuttings also can influence rooting potential. Higher CO₂ levels in the range of 1800 to 200 ppm supplied for 12 hours per day have increased rooting. Carbonated mist has been used with similar results. Also, increasing the medium temperature by the use of hot water or electric heating cables has speeded root development and in some cases increased the range of plants propagated by cuttings.

The most recent development which may, in fact, have a substantial impact and wide-range implications in plant propagation has been the development of the technique of tissue, or meristem, culture. Cultures of tissues from carrot, tobacco and Jerusalem artichoke have been grown for a number of years. The first practical application of the technique on a wide scale has been the propagation of orchid species by excising the meristem and growing them on a defined medium. Other tissues from herbaceous and woody plants have been

successfully cultured. However, one of the biggest problems with tissue culture and one which is very similar to the problems incurred in root initiation is the inability to cause the tissues to differentiate into shoots and roots from the callus stage. We are all familiar with cuttings which may produce abundant callus but yet stubbornly refuse to initiate primordia. Although in tissue culture, roots frequently do form in the callus, it is considerably more difficult to obtain shoot differentiation. One of the great problems, therefore, facing researchers in the physiology of plant propagation—and it is perhaps one of the great unsolved problems of biology—is the understanding of what regulates the differentiation of a group of cells into a recognizable plant organ. Since all cells contain the information needed to develop into a total plant, the question is: why is only a small portion of the information available at any one time, and what regulates what information will be made available to direct the development of a group of cells into a shoot or root at a particular point in time? As we learn more about the regulation of differentiation we will be better able to regulate root formation in cuttings and also expand the use of tissue culture as a commercial practice in plant propagation.

MODERATOR FURUTA: Thank you, Charlie. I think we will take one or two questions now if you have any at this time.

VOICE: Is there a method or formula for determining the minimum number of foot candles necessary for root initiation?

CHARLES HESS: Of course you can't answer that in a general way, because it varies from plant to plant. Plants such as dogwoods, for example, which are an understory plant, will tolerate less light—will reach maximum photosynthesis under less light intensity than say, a plant that grows under full light intensity. Now that I say you can't do it, let me tell you what, as a general rule, we use. Saran cloth, at about a level of 20% shade, seems to work out all right. We used to use cheese cloth, but Saran is more stable; it won't break down as readily. This use of shade has a number of advantages. It does, apparently, allow enough light through to give ample photosynthesis to a wide range of plant materials. It also reduces certain problems if you're outside, such as wind affecting the mist bed. Those are the two things that I think of right off hand. I would say, even in New Jersey, where we don't have all the sunshine that you have here, that outside it is advisable to use some degree of shade. The other advantage that I think of in using shade is to reduce the frequency of mist applications. Our goal is to try to just keep the leaf with a film of water on it. Any more than that really is probably a surplus and can cause problems, as far as lowering the rooting medium temperature, water-logging of the medium and so forth. So if you use this combination of mist and a light

shade, you can, perhaps, get a little closer to the optimum set of environmental conditions.

FRANCES SPAULDING: I wonder if you have any idea where ethylene fits into the picture of root initiation.

CHARLES HESS: It had been observed for many years, before ethylene became a very fashionable thing to study in plant physiology, that ethylene did stimulate root initiation. A good example is tomatoes. If tomatoes are exposed to a low quantity of ethylene, then lots of roots form up and down the stems; there are people who are suggesting that the effect of an auxin in stimulating root initiation is in injuring the tissues a little bit so that they will produce ethylene. It's ethylene that's doing the job as far as root initiation is concerned. At this point, I'm not really prepared to agree with that — but I can't disagree with it, either.

MODERATOR FURUTA: Our next topic will be, as presented in the program, Staking, Pruning and Spacing. And with this I think the speakers have a wide latitude; you could go almost anywhere. So I should like to introduce at this time two gentlemen who need no introduction to this group, Dr. Andy Leiser and Dr. Richard Harris.

TREE TRUNK DEVELOPMENT:

INFLUENCE OF STAKING AND PRUNING¹

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Abstract. Trunk development of young container-grown trees was strongly influenced by pruning and staking practices. Trees were produced which were able to stand without support when planted in the landscape. This was done by eliminating stakes, leaving lateral branches on the trunk and spacing plants so their tops were free to move. Even though rigidly-staked trees

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