HORMONES AND PROPAGATION

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INTRODUCTION

That plant hormones have a marked effect on the rooting of cuttings has been recognized for some 40 years, since it was first demonstrated that apption of the auxin-type hormone, indolyl-3-acetic acid (IAA), will stimulate rooting in stem cuttings of variousspecies. For many years the *auxins* (which chemically are indole derivatives) were the only known natural plant hormones, but in the last 15 to 20 years it has become recognized that there are several other groups of natural plant hormones, including the *gibberellins* and *cytokinins*, which are growth promoting substances, *abscisic acid*, which often behaves as a growth inhibitor, and *ethylene*, the first known growh regulating substance which is a gas. These various types of hormone are very different from each other chemically. Thus, the gibberellins are terpenoid-like compounds with a fairly complex structure, while the cytokicns are purines, being substituted adenines or closely related substances. There are now about 40 known naturally-vcurring gibberellins, but the number of identified natural cytokinins does not exceed about 10.

As well as the natural hormones there is a wide range of synthetic substances which have hormone activity. Thus, there are quite a number of substances which will stimulate the rooting of cuttings, including the well-known compound indolylbutyric acid (IBA). There is also a wide range of substances with cytokinin activity, including a number of substituted ureas. However, so far the gibberellins have proved too difficult to synthesize and only the naturally-occurring substances are available at present.

Each of these types of hormone has a wide spectrum of effects, and while each type of hormone may have some effects which are unique, in many cases two or more types of hormone may affect the same process, e.g. stem growth. For example, it has been known for many years that a developing bud on a woody shoot will stimulate division in the cambium in the stem below it, and it was later found that the stimulating effect of a bud can be partly replaced by applying IAA to disbudded twigs. Since developing buds themselves produce IAA which is transported downwards in the stem it is very probably that the stimulating effect of an expanding bud on the cambium is due to the natural auxin which it produces.

When gibberellins were recognized to be natural plant hormones, it was found that gibberellic acid would also stimulate

division of the cambium to some extent but, whereas when IAA alone is applied, the cells derived from the cambium form xylem (wood), when GA is applied they form phloem (bark). However, when both IAA and GA are applied together their combined effect on cambial division is very much greater than with either alone, and active xylem and phloem production takes place. Since developing buds produce gibberellins as well as auxins, it seems likely that the natural cambial stimulus produced by buds includes both types of hormone. These findings suggest that it may be useful to apply a mixture of IAA and GA when making a graft, in order to stimulate cambial activity and improve the likelihood of a successful graft union.

The other growth-promoting type of hormone, the cytokinins, also stimulates cell-division. There are a number of cases where cytokinins have been reported to stimulate cambial division, but it has no effect with some woody species. However, it also stimulates the disorganized type of cell division which leads to callus formation. Moreover, when cytokinin and auxin are applied together to pieces of tobacco pith in sterile culture, the effect depends upon the relative concentrations of the wo hormones: when the concentration of auxin is high and that of cytokinin is low then the pith cells will proliferate and regenerate roots; but when the level of cytokinin is high and that of IAA is low, then buds are regenerated. This finding probably has some significance for propagation by root cuttings, since this depends upon the cutting regenerating not only roots from the "lower" bud, but also buds from the "upper" end. The eminent French plant physiologist, the late Dr. Jean Nitsch, studied the hormone changes occurring in pieces of chicory root during regeneration, and found that they passed through a phase in which relatively high concentrations of the natural cytokinin accumulated at the top, and likewise auxins at the base of the cutting, before bud or root regeneration had occurred. Thus, the natural pattern of regeneration in root cuttings may be understood in terms of the distribution of natural auxins and cytokinins which occurs within them.

The other two types of natural hormones, abscisic acid (ABA) and ethylene, are important for propagation problems mostly indirectly, through their role in seed and bud dormancy. However, it has been reported that ABA will promote rooting of cuttings in a few instances, such as in adult ivy [Hedera helix]. In this latter case it is thought that the ABA acts by inhibiting the effects of the natural gibberellins, which are inhibitory to rooting in some species. Thus, the effect of ABA is apparently to inhibit an inhibitor of rooting! Ethylene has marked effects upon germination in a number of species, stimulating the germination of dormant seeds in some cases and inhibiting it in others.

It is characteristic of hormones that they are produced in one part of the plant and have effects in some other part. Thus auxins appear to be produced at the growing points of the shoot and to move downwards in the stem from there. Cytokinins are apparently formed in the root tips and move up into the shoot with the sap. Gibberellins appear to be formed in the tips of both shoots and roots.

HORMONES AND THE ROOTING OF SOFTWOOD CUTTINGS

Cuttings of many herbaceous plants can, of course, be very easily rooted without any special hormone treatment. This does not mean, however, that hormones are not involved in t. j rooting of such plants, but simply that the natural hormone iↄ.els are sufficiently high to ensure "natural" rooting. Young developing leaves, whether in the terminal bud of an actively growing leafy shoot, or the expanding resting buds of a woody plant in the spring, contain high levels of natural auxin, presumably IAA, which moves predominantly downwards in the stem. When a cutting is taken, the downward-moving auxin will accumulate at the base of the cutting and presumably reach a high enough level to stimulate root initiation. When an external source of auxin, such as IBA, is applied it is simply stimulating and augmenting the natural process. No doubt the effectiveness of layering is partly due to the fact that the cut in the stem blocks the downward movement of natural auxin and leads to the accumulation in the downward-pointing part of the layered stem.

What we have said about the rooting of cuttings of herbaceous plants no doubt applies equally to "softwood" cuttings of woody plants, i.e. the auxin produced in the young developing leaves at the growing point moves downwards and accumulates at the base of the stem. However, it is well-known to horticulturalists that, in many species, as the season progresses there is a decline in the rooting responses of softwood cuttings.

What is the reason for this seasonal variation in rooting ability? One possible explanation might be that many woody plants, in contrast to herbaceous ones, cease extension growth and set a terminal resting-bud by September, if not earlier. It is known that when the growing point ceases to produce actively-growing young leaves, its level of auxin production falls drastically. Thus, it would seem quite possible that the decline in rooting ability in the later part of the growing season, which is observed with softwood cuttings of many woody species, is due to the fall in auxin production when a resting bud is "set".

We have investigated this question in *Populus* x *robusta* (11). We compared the rooting abilities of softwood cuttings which had either (a) an active growing point, (b) a dormant terminal

resting bud, or (c) had had their active growing points removed leaving only mature leaves. Somewhat to our surprise the differences in rooting response were relatively small (Table 1), although the cuttings with an active growing point gave the highest percentage of rooted cuttings and the highest number of roots. Moreover, further studies indicated that dormant leafy cuttings contained no less auxin than actively growing ones. Although mature leaves produce less auxin than young expanding ones, it would appear that even fully expanded, mature leaves of poplar produce sufficient auxin to allow good rooting ability in the absence of an active growing point. Whether the same situation applies to leafy cuttings of other woody plants is not known, but it has been reported recently that removing the growing point of dahlia cuttings actually improves the rooting response (2), apparently because the growing point competes with the rooting region of the stem for nutrients and so removal of the shoot tip makes more nutrients available for root formation.

Apart from "internal" factors within the shoot itself, such as the presence or absence of an active growing point, there are various environmental factors which affect rooting and which may account for seasonal trends in rooting ability. An important environmental factor is the seasonal variation in day length. It is well-known that many woody plants are sensitive to daylength and tend to form resting buds as the days get shorter in the fall, but from our work with poplar described above it would appear that the seasonal decline in daylength cannot be attributed solely to the cessation of extension growth later in the season. However, there are a number of reports that daylength may affect the rooting ability of leafy cuttings, quite apart from its effects upon extension growth (see Smith and Wareing (10), for the relevant literature). Thus, Waxman (13) showed that if the source plants of Cornus florida cuttings were kept under either 15 hour or 9 hour days for 4 months prior to taking cuttings, the "long day" cuttings rooted readily whereas the "short day" ones failed to root. Similar results were obtained with Populus x robusta (9). On the other hand, in a survey of 26 woody ornamental species, Baker and Link (1) found that the majority of species did not show any significant response to daylength treatments.

We have investigated the effects of daylength on the rooting of softwood cuttings of Populus x robusta taken at monthly intervals from 26 May to 22 September, from stooled plants growing in the open. The cuttings were placed in a rooting medium and maintained under either 18 hour or 9 hour days. We found that the percentage of cuttings rooted and the average number of roots formed were both higher under long days than under short days. However, there was a steady decline in the rooting ability of even the "long day" cuttings taken on successive dates.

Moreover, this decline was accompanied by a corresponding decline in the levels of auxins extracted from the cuttings.

Howard (6, 7) has shown that in apple and plum cuttings there is a marked decline in the rooting ability in the early winter (Nov - Dec) and that this effect cannot be overcome by applying IBA. Thus, it appears that the decline in rooting response in the fall is not simply due to a decline in the levels of endogenous auxin. It is possible that in deciduous species this decline in rooting ability in the late autumn — early winter period may be due to leaf-fall. Certainly, we have found that leafy cuttings of forsythia taken in October and placed in mist formed many more roots than corresponding defoliated cuttings. If the autumn decline in rooting response is due to leaf-fall, then the stimulating effect of leaves on rooting must be due to the supply of substances other than auxins, since otherwise it should be possible to obtain a good rooting response at that time by applying IA, whereas we have seen above that this is not the case. The cause of this seasonal decline in rooting ability in the autumn merits further study.

HARDWOOD CUTTINGS

There is a marked seasonal periodicity in the rooting of hardwood cuttings. Thus, cuttings taken in late autumn — early winter (Oct - Dec) frequently show a very poor rooting response, whereas from January onwards the rooting response improves steadily (6, 7). It is also known that the rooting of hardwood cuttings in the spring is stimulated by the presence of an expanding bud, and that disbudding markedly reduces the rooting response (3, 8). By contrast, disbudding hardwood cuttings in October either has little effect or may actually increase rooting. Thus, it would appear that it is only non-dormant buds which stimulate rooting.

Since the buds of many temperate woody species require a period of chilling to overcome the dormancy of their buds, it seemed likely that the improved rooting response as the winter progresses may be due to the fact that the buds gradually lose their dormancy as their chilling requirement is met. We, therefore, compared the rooting responses of chilled and unchilled cuttings of *Populus x robusta* and found that rooting was very much greater in the chilled than in the unchilled cuttings (10). We also studied the rooting responses of chilled and unchilled *disbudded* cuttings and found that disbudded chilled cuttings formed many fewer roots than cuttings with chilled buds. However, chilling improved the rooting response even of the disbudded cuttings, suggesting that chilling has a promotive effect on rooting over and above its effects upon the dormancy of the buds,

and that chilling may, in fact, enhance the rooting ability of stem tissues directly and independently of any effects on the buds. Parallel studies on the levels of the endogenous auxins in relation to chilling and disbudding indicated that there were higher levels of auxins in chilled than in unchilled poplar cuttings, but disbudding appeared to have little effect on the auxin levels. However, it is possible that some auxin had already been produced by the buds on the parent plant, before the cuttings were taken and disbudded.

Not only does the presence of expanding buds have a stimulating effect on rooting, but also upon cambial activity. Thus, disbudded shoots of poplar show little renewal of cambial activity in the spring, but the effects of the buds can be replaced by applying IAA and GA (see above). Thus, so far as the cambium is concerned, the stimulatory effects of the buds in the spring appears to be mediated through the hormones which they produce. It is reasonable to think, therefore, that the stimulatory effect of non-dormant buds on rooting is also attributable to the hormones they produce.

Howard (7) reported that disbudding had little effect on the progressive increase in rooting response of Myrobalm plum cuttings during the winter, and concluded that "changes in the capacity of the cuttings to root between autumn and spring are not related to the state of activity of the buds on the cutting as influenced by the winter environment". However, it should be noted that Howard applied IBA to the bases of all cuttings in his experiments, and this treatment would obscure any effect of disbudding mediated through reduced endogenous auxin levels. Nevertheless, his results are interesting in that they show that there is a progressive increase throughout the winter in the rooting response of stem tissue to applied auxin. Thus, in addition to the effects of chilling on the dormancy of the buds, chilling appears to increase the responsiveness of the stem tissues to auxin, a conclusion which is consistent with the effect of chilling on disbudded poplar cuttings observed by Smith and Wareing (see above).

As well as increasing the production of auxins by the buds, chilling also leads to an increase in content of cytokinins in Populus x robusta shoots (5), and of gibberellins in other species (12). The extent to which changes in hormones other than auxins are involved in rooting responses needs further study.

CONCLUSIONS

Although it is clear that natural plant hormones are closely involved in several aspects of plant propagation, it is also clear that our understanding of their role is very incomplete in many

ways. Not only do we need more intensive studies on the function of the well-known auxin-type hormones, but we also need to find out how the recently discovered gibberellins and cytokinins are involved, either directly or indirectly. Moreover, it is becoming increasingly apparent that the rooting of cuttings involves other endogenous substances in addition to hormones, such as the terpenoid factors involved in the rooting of ivy cuttings (4), and we need to know how such factors interact with hormones in controlling root initiation, cambial activity and other processes bearing on vegetative propagation.

Table 1. Effect of resting bud and decapitation on rooting of leafy cuttings of *Populus x robusta*. From Smith and Wareing, (11)

Type of Cutting	Mean No roots per cutting.	Percentage rooting	Mean No leaves per cutting	Root/leaf number ratio
A With dormant apices	9 2	96 4	4 6	1 98
B With active growing points	8 1	75.0	4 0	2 00
C Decapitated	7 6	82.1	2.9	2 64

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