COMPARATIVE ACTIVITY OF ROOT-INDUCING SUBSTANCES AND METHODS FOR TREATING CUTTINGS

A. E. Hitchcock and P. W. Zimmerman

INTRODUCTION

Experimental work extending over several years has resulted in the development of a number of methods for treating cuttings with root-inducing substances. Although each method has proved effective at least on certain plants, there has been a tendency to work toward more simplified procedures for use in practice. Modified methods in which the duration of treatment is reduced to a minimum of a few seconds simplifies application of the root-inducing substances, but introduces complications associated with the use of concentrations 10 to 1,000 times higher than those used for the standard 24-hour treatment (3).

Results with powder preparations are complicated by the fact that the carrier most commonly used (talc) showed some activity itself and thus consistently caused better rooting than was obtained on non-treated or tap water controls. In addition, the effectiveness of powder preparations was noticeably dependent upon the mechanical fineness of the powder. For this reason the results of different workers may not always agree unless strictly comparable preparations are used.

It is the purpose of the present paper to present the results of comparative tests making use of several methods of subjecting cuttings to the action of root-inducing substances. Results with powder preparations (0.5 to 50 mg./g.) and with solution concentrates (1 to 20 mg./cc.) showed that different dosages were required for different kinds and types of cuttings just as in the case of the standard 24-hour treatment with 1 to 80 mg. per liter. Whichever method is used, of the three just mentioned, the root-inducing substance must first be in solution before it can enter the tissue. It has not been determined how vapors enter the tissue. The powder preparations were more effective when the basal ends were moistened before being dipped into the powder.

MATERIALS AND METHODS

Indolebutyric, naphthaleneacetic, and indoleacetic acids and their potassium salts were the principal root-inducing substances used. In one test the methyl esters of naphthaleneacetic and indolebutyric acids were applied as vapors to the cuttings for a period of 30 minutes, in a bell jar, and then the cuttings were planted as described below. The species of cuttings used are listed in Table I. Other species treated with powders only, appear in the text.

Standard solution immersion method. Basal ends of cuttings were immersed for 24 hours in water containing the root-inducing substance in concentrations of 1 to 80 mg./l. (milligrams per liter of water). Control lots were immersed in tap water for the same period. After treatment the cuttings were planted in greenhouse benches or in cold frames.

Concentrated solution dip method. Solutions containing the root-inducing substance in concentrations of 1 to 20 mg./cc. (milligrams per cubic centimeter) were used in these tests. The solvent was either water, alcohol-water mixtures, or diethyleneglycol. Basal ends of the cuttings were dipped into the solution to a depth of about one-half inch. Control cuttings were similarly dipped into tap water. After treatment the cuttings were planted in greenhouse benches or in cold frames.

Powder dip method. Basal ends of cuttings were dipped to a depth of about one-half inch in powders containing the root-inducing substance. The method of dipping had to be modified slightly according to the number of cuttings which were treated simultaneously. Thus to secure uniform coating of the basal part of the stem it was necessary to stir the immersed parts to the extent of one to two revolutions in cases where a group of cuttings was treated. The cuttings were then tapped gently on the rim of the container in order to remove excess powder. Since in preliminary tests the dipping of cuttings with dry stems proved relatively ineffective for most species, the final method adopted was to wet the basal ends of the cuttings just before dipping them into the test powders. Control cuttings were similarly wetted and then dipped in talc to which a root-inducing substance had not been added.

Two types of powders were used in the tests. In one case a commercial grade of talc (Eimer and Amend) and the root-inducing substance were mixed in the proportions of 1, 4, 10, 25, and 50 mg. of the root-inducing substance per gram of talc. All of the root-inducing substances mentioned in the first part of this section were obtained from Merck & Co. Inc., Rahway, New Jersey. A second series of powders prepared by Merck & Co. Inc., contained the root-inducing substances in various concentrations ranging from 1 to 35 mg./g. In order to avoid confusion, exactly the same concentrations in the two series are not reported in this paper, even though all concentrations were used originally. Thus concentrations of 2, 5, 6, 12, 18, and 35 mg./g. have been used to designate powders prepared by Merck & Co. Inc. Merck powders tested, but for which specific data (with one exception) are not presented, were as follows: 0.5, 1, 3, 8, 10, and 16 mg./g. The one exception is the 1 mg./g. powder which was used on the Briarcliff rose (Fig. 1 A) and chrysanthemum (Table II). The Merck powders appeared to be of a finer texture than the powders which were prepared in our laboratory.

As in the case of the other methods, the cuttings were planted immediately after treatment with the test powders.

RESULTS

CONCENTRATION REQUIREMENTS ACCORDING TO THE METHOD OF TREATING THE CUTTINGS

Data in Table I show that the optimum concentration of indolebutyric acid varied with the species regardless of whether the root-inducing substance was applied as a relatively dilute solution, as a concentrated solution, or as a powder. Species such as the Briarcliff rose (Fig. 1 A and Table III), begonia, ivy, and chrysanthemum (Table II) responded to treatment with relatively low concentrations with all three methods. Likewise, certain species responding best to relatively high concentrations with one method also required relatively high concentrations with the other two methods. Some of the species falling in the latter class were *Thuja occidentalis* var. *robusta*, *Chamaecyparis obtusa* var. *filiformis*, *Rhododendron* (Fig. 2, A and C), *Tsuga canadensis* (Fig. 3 A), *Abies veitchii*, *Syringa vulgaris* (Table IV and Fig. 2 B), *Picea glauca* var. *conica*, and *Camellia* (Fig. 2 D). Species requiring intermediate concentrations were carnation (Fig. 1 B), dahlia, *Prunus*, elm, *Celastrus*, and *Caragana*.

Concentrated solutions and powders containing indolebutyric acid were about equally effective on a weight basis of root-inducing substance per gram of carrier (Table I), but the latter values are 10 to 1,000 times the concentrations (by weight) which induced equivalent rooting when applied according to the standard immersion method of treatment. The concentrated solution dip method was generally more critical than the other two methods but the results obtained with the former were particularly satisfactory in the case of species difficult to root such as the apple (Fig. 2 E), hemlock (Fig. 3 A), rhododendron (Fig. 2, A and C), and many of the evergreens listed in Table I.

Although the response of *Pachysandra* (Fig. 1 C) and *Hibiscus* (Fig. 1 D) indicates tolerance to a relatively wide range of concentrations, it is to be noted that concentrations of 18 to 35 mg./g. caused toxic effects consisting of the production of an excessively large number of roots, or only a few roots, failure to root, excessive proliferation, the retardation or inhibition of bud growth, or killing of the lower part of the stem. Thus for *Pachysandra* the optimum concentration was between 5 and 12 mg./g. and for *Hibiscus* between 6 and 18 mg./g. Since lower concentrations were effective in producing more roots than on control cuttings, these two species represent tolerant types. Most of the other species listed in Table I were not so tolerant.

The same correlation between the average number of roots per cutting and concentration as was previously reported for the standard immersion method of treatment (4) was also found to hold for the powder and concentrated solution dip methods (Tables I, V, and VI and Fig. 3). It is

TABLE I

EQUIVALENT CONCENTRATIONS OF INDOLEBUTYRIC ACID FOR ROOTING CUTTINGS BY THREE DIFFERENT METHODS

Name of plant	Time of year treated	Concentration of indolebutyric acid*			
		Mg./l. (24 hrs.)	Mg./cc. (dip)	Mg./g.** (dip)	Mg./g.† (dip)
Abies veitchii Murr. (fir)	Jan.	60–80	10–20	10–50	2–12
Acer palmatum Thunb. (Japanese maple)	May–June	10–40	—	—	2–5
Actinidia arguta Miq.	Dec.–Jan.	20–40	1–4	4–10	—
Begonia sp. var. Marjorie Gibbs	Dec.	1–5	—	—	1–2
Begonia semperflorens Link. & Otto	Nov.	1–5	—	—	1–2
Camellia japonica L. var. *chandleri elegans*	Jan.	60	4–10	25–50	—
Caragana boissii Lam.	May	10	4	—	2–5
Carya pecan Aschers & Graebn. (pecan)	Apr.	40	—	—	2–12
Catalpa sp.	Apr.	—	4	—	5–12
Celastrus articulatus Thunb. var. *punctatus* (bittersweet)	Nov.–Apr.	40	1–4	4–25	1–2
Cerastium sp.	Apr.	2–5	—	—	1–5
Chamaecyparis obtusa Sieb. & Zucc. var. *lutea nova*	Apr.	40–60	—	—	5–12
Chamaecyparis obtusa Sieb. & Zucc. var. *filiformis*	Apr.	40–60	—	—	5–12
Chamaecyparis pisifera Sieb. & Zucc. var. *plumosa aurea*	Apr.	40–60	—	—	5–12
Chamaecyparis pisifera Sieb. & Zucc. var. *filifera aurea*	Nov.	60	—	10–25	5–12
Chilopsis linearis D.C. (desert willow)	Jan.	60	4	10–25	—
Chrysanthemum sp.	Aug.–May	1–10	—	4–10	1–5
Crassula sp.	May	40	—	—	2
Cryptomeria japonica D. Don	Dec.	40–80	4–10	10–25	—
Dahlia variabilis Desf. (dahlia)	Apr.	20	4	—	2–5
Dianthus caryophyllus L. (carnation, English varieties)	Nov.–Mar.	5–10	—	4–10	1–5
Diervilla hybrida Dipp. var. Mme. Billard (weigela)	Oct.	5–10	4	—	1–2
Euonymus radicans Sieb.	Nov.–Apr.	5–10	1–4	—	1–5
Euphorbia pulcherrima Willd. (poinsettia)	Apr.	2–10	—	—	2
Gossypium hirsutum L. (cotton)	Mar.	—	—	—	5–12
Hedera helix L. (ivy)	Nov.	1–5	1	1–4	1–2
Heliotropium sp. (heliotrope)	Jan.–Feb.	10	—	—	1–2
Hibiscus syriacus L. (althea)	Oct.–Feb.	40–60	4–10	4–50	2–18
Ilex aquifolium L. (English holly)	May	20–40	—	—	5
Ilex opaca Ait. (American holly)	Nov.	40	4–10	10–50	—
Juniperus chinensis L.	Mar.	60	4	—	2–5
Juniperus chinensis L. var. *pfitzeriana* Spaeth.	Dec.–Jan.	60–80	4–10	10–25	2–12
Juniperus communis L. var. *montana* Ait.	Apr.	40–60	—	—	5–12
Juniperus conferta	Apr.	40	—	—	2–12
Juniperus virginiana L. var. *tripartitia* R. Smith	Apr.	40–60	—	—	5
Kolkwitzia amabilis Graebn.	June	20	—	—	12
Ligustrum ovalifolium Hassk. (privet)	Oct.–Nov.	40	4–10	10–50	—
Ligustrum ovalifolium Hassk. (privet)	May	80	10	—	2–12
Pachysandra terminalis Sieb. & Zucc.	Feb.	20–40	—	—	5–12
Picea abies (L.) Karst. (spruce)	Jan.–Mar.	20–40	—	—	2–5
Picea glauca Voss. var. *conica* (spruce)	Mar.	40–60	4	—	2–12
Prunus cerasifera Ehrh. var. *woodii*	May	—	4	—	2–5
Prunus persica Sieb. & Zucc. (Elberta peach)	Nov.	20–40	1–4	10–25	—
Prunus serrulata Lindl.	June	—	4	—	1–2

TABLE I—*Continued*

Name of plant	Time of year treated	Concentration of indolebutyric acid*			
		Mg./l. (24 hrs.)	Mg./cc. (dip)	Mg./g.** (dip)	Mg./g.† (dip)
Pyrus malus L. (Grimes Golden apple)	Nov.	40	4–10	10–25	—
Pyrus malus L. (Rhode Island Greening apple)	Nov.	40	4–10	10–25	—
Rhododendron sp. (*catawbiense* hybrids)	Dec.	40–80	10–20	10–50	5–12
Rhododendron sp. var. Caractacus	Apr.	—	10–20	—	5–12
Rosa sp. var. Briarcliff (rose)	Sept.–Mar.	1–2	0.3–1	—	1–2
Rosa sp. var. Crimson Rambler (rose)	May	1–5	1–4	—	1–2
Styrax americana Lam.	Dec.	—	4	25	—
Syringa emodi Wall. (lilac)	May	40	4	—	2–12
Syringa vulgaris L. (lilac)	Apr.–May	20–60	4–10	—	2–12
Taxus cuspidata Sieb. & Zucc. (yew)	Oct.–Feb.	60	4–10	10–50	2–5
Thuja occidentalis L. var. *ellwangeriana* Beissn.	Apr.	40	4	—	—
Thuja occidentalis L. var. *globosa nana*	Nov.–Apr.	20–60	4	—	2–12
Thuja occidentalis L. var. *robusta* Carr.	Nov.	60	4–10	25–50	5–12
Thuja occidentalis L. var. *spiralis* Hort.	Jan.–Apr.	60	4	4–25	2–12
Tsuga canadensis Carr. (hemlock)	Dec.–Feb.	40–60	4–20	10–50	5
Ulmus americana L. (elm)	Mar.	10–20	2–4	—	2–5
Vaccinium corymbosum L. var. Rubel (blueberry)	June–July	20–40	—	—	2–5
Viburnum carlesii Hemsl.	June	—	4	—	2
Vitis sp. (Concord grape)	Oct.	40–60	4–10	—	—
Vitis sp. (Concord grape)	Nov.	40	4–10	10–25	2–10

* Equivalent concentration values are described under "Materials and Methods."
** Commercial grades of talc and acid mixed in proportions indicated.
† Specially prepared finely ground mixtures of talc and indolebutyric acid.

believed that this relationship is a far more reliable criterion of the relative activity of different root-inducing substances than the percentage of cuttings rooted, since marked differences may be shown even when all cuttings (including controls) are rooted. For example, the marked difference in the rooting response illustrated for *Euonymus* in Figure 2 G would be entirely lost if the results were expressed in terms of the percentage of cuttings rooted. On the latter basis the poorly rooted dry controls and the 5 mg./g. lots which show 100 per cent rooting (Fig. 2 G, lower row) would be rated equivalent to the excellently rooted 5 mg./g. wet-treated lot appearing on the right in the upper row.

Concentration requirements varied not only with the species (Table I) but also with the relative age of the cutting with particular reference to the degree of hardness where the basal cut was made. Thus the optimum concentrations for succulent tip cuttings of chrysanthemum were 1 to 5 mg./l. and 0.5 to 1 mg./g., whereas for cuttings with harder stems the optimum concentrations were 5 to 10 mg./l. and 2 to 5 mg./g. (Table II). Both types of cuttings may be secured from the same plant or from shoots on plants grown under different conditions, as for example at different temperatures or at different times of the year. In contrast to chrysanthe-

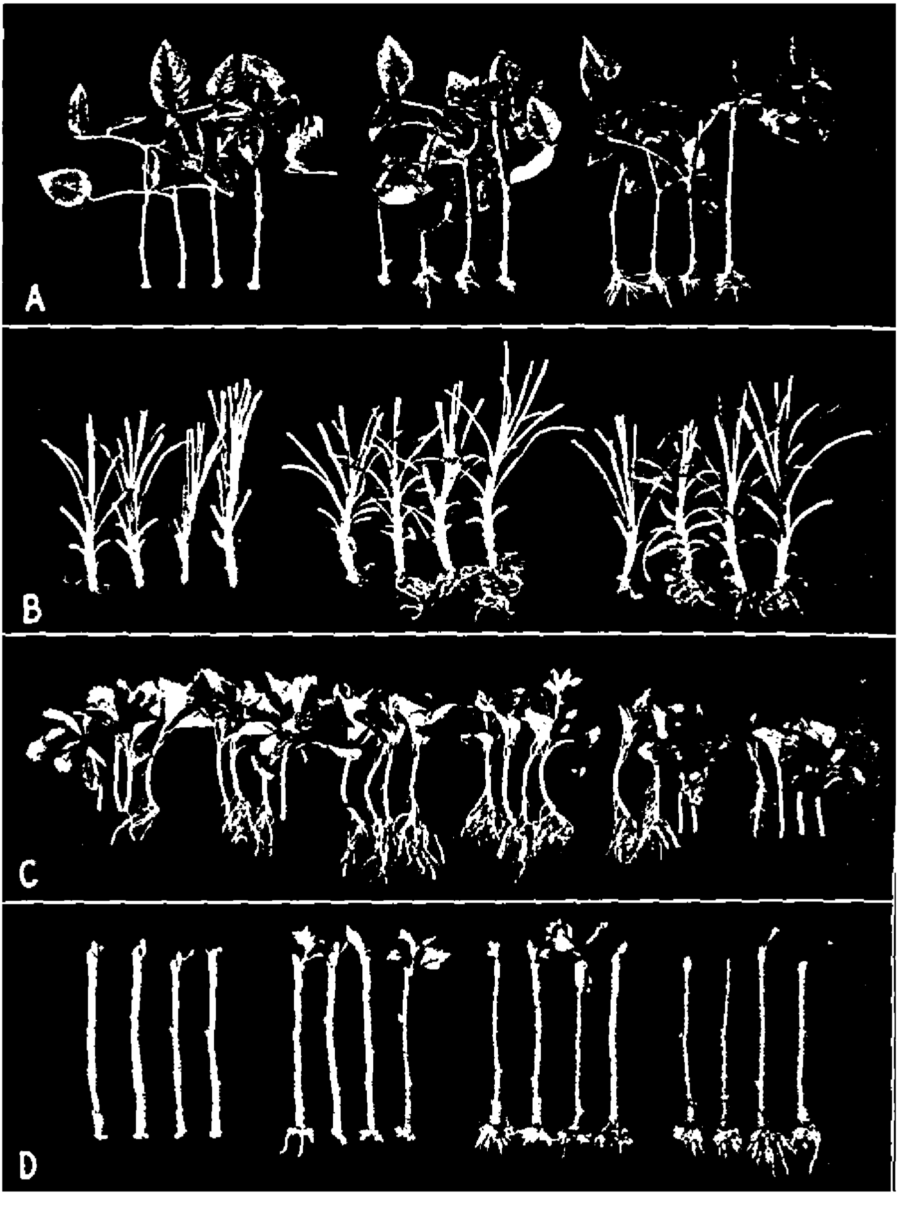

FIGURE 1. Concentration requirements (indolebutyric acid powder) for different species. A. Briarcliff rose (*left to right*): talc control, 1 and 2 mg./g. respectively. B. Carnation var. Pelargonium (left to right): talc control, 2 and 5 mg./g. respectively. C. *Pachysandra terminalis* (left to right): talc control, 2, 5, 12, 18, 35 mg./g. D. *Hibiscus syriacus* (left to right): talc control, 6, 18, 35 mg./g.

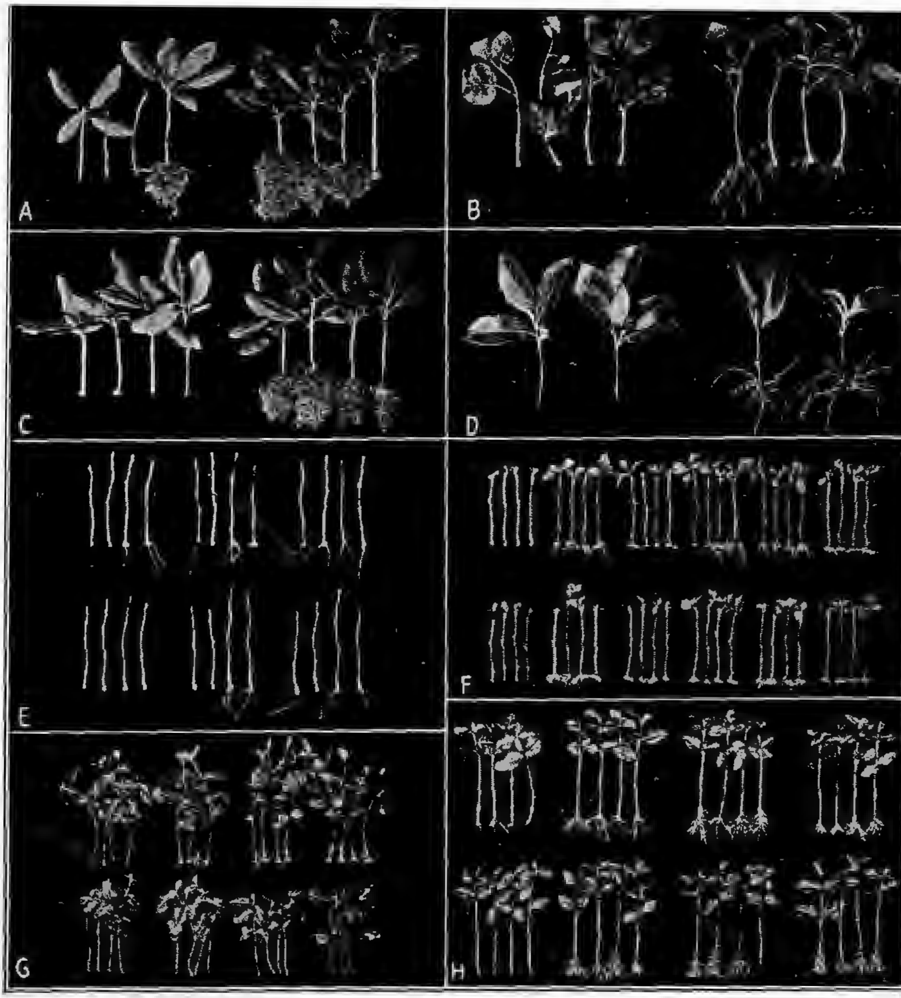

FIGURE 2. Response of cuttings to indolebutyric acid or the K-salt. Key: mg./g. signifies powder dip; mg./cc., solution dip; and mg./l., 24-hour immersion method. A. Rhododendron (left to right): check, 25 mg./g. acid. B. Lilac var. Arthur Wm. Paul (left to right): check, 12 mg./g. acid. C. Rhododendron (left to right): check, 10 mg./cc. K-salt. D. *Camellia japonica* var. *chandleri elegans* (left to right): two checks, 10 mg./cc., 25 mg./g. acid. E. Grimes Golden apple. (Upper) Acid solution (left to right): check, 4 and 10 mg./cc. respectively. (Lower) Acid powder (left to right): check, 10 and 25 mg./g. respectively. F. *Hibiscus syriacus*. (Upper) Bases washed at different intervals after treatment with 25 mg./g. acid. (Lower) Washed, then retreated. Left to right (both rows): check; treated but not washed; washed after 2 hours, 1 day, 3 days, 7 days respectively. G. *Euonymus radicans*. (Upper) Wet cuttings. (Lower) Dry cuttings. Left to right: check; 1, 2, 5 mg./g. acid respectively. H. *Euonymus radicans*. (Upper) Acid. (Lower) K-salt. Left to right (both rows): check; 4 and 10 mg./g.; 20 mg./l. (24 hours).

TABLE II

ROOTING RESPONSE OF CHRYSANTHEMUM CUTTINGS TREATED WITH INDOLEBUTYRIC ACID APPLIED IN DIFFERENT FORMS. AVERAGE NUMBER OF ROOTS PER CUTTING

Variety	Date treated	Date of root count	Concentration of indolebutyric acid								
			Mg./g. (dip)				4 mg. per cc. (dip)	Mg./l. (24 hrs.)			
			0*	1	2	5		0**	1.25	2.5	5
Rowena	Apr. 6	Apr. 17	4	—	9	—					
Valencia	Apr. 6	Apr. 17	10	49	42	47					
Melba	Apr. 6	Apr. 17	6	—	25	14		1	20	—	—
Heldaberrs	Apr. 6	Apr. 17	6	—	20	31					
Teton	Apr. 6	Apr. 17	6	—	20	30					
Harvard Red	Apr. 6	Apr. 17	2	6	14	13		1	—	5	—
Harvard Red	Apr. 14	Apr. 25	13	20	13	17	9	6	—	15	19
Peggy Ann Hoover	Apr. 6	Apr. 19	5	8	5	8		3	12	9	—
Peggy Ann Hoover	Apr. 14	Apr. 25	8	—	16	21		4	—	11	14
Peggy Ann Hoover	May 26	June 8	10	—	14†	—	8	3	—	—	10
Hilda Bergen	Apr. 14	Apr. 25	21	—	59	91					
Red Bird	Apr. 20	May 4	13	—	40	44		7	—	39	45
Red Bird	May 26	June 6	20	—	121†		47	11			140
Nugget	Apr. 26	May 9	8	—	23	25		13		18	25
Nugget	May 26	June 8	15	—	35†		30†	2	—	—	22
Yvalda	May 26	June 8	28	—	45†		0†	13	—	—	47

* Finely ground control talc.

** Tap water control, 24-hour treatment.

† Some or all cuttings definitely injured, as evidenced by excessive proliferation, discoloration, or killing of lower part of stem, or noticeable retardation of root growth.

mum, the tip cuttings of *Hibiscus syriacus* required higher concentrations than cuttings made from the remaining portion of the shoot. These differences are well-known and have been previously discussed in connection with solution treatments (4). The relative age of the cutting is thus an important limiting factor in determining the rooting response of cuttings treated with powders and with concentrated solutions.

The time of year treatments were administered also proved to be an important limiting factor in the response of cuttings to root-inducing substances. As previously reported for the named varieties of *Syringa*

TABLE III

REDUCTION IN NUMBER OF ROOTS CAUSED BY COATING BARK ON LOWER PART OF BRIARCLIFF ROSE CUTTINGS WITH PARAFFIN BEFORE TREATMENT WITH THE ROOT-INDUCING SUBSTANCE

Treatment with indolebutyric acid	Bark not coated	Bark coated*
24-hour treatment 2.5 mg./l.	39	17
Solution dip method 1 mg./cc.	38	13
Powder dip method 4 mg./g.**	11	2
Column totals	88	32

* Root-inducing substance in direct contact only with basal cut surface.

** Mixture not finely powdered.

vulgaris which responded to solution treatment only when taken the latter part of April or during May (6), the same results were obtained during 1939 with the powder treatments. Even though young shoots of the lilac taken during May are difficult to maintain in good condition in the rooting medium, they tolerate and require relatively high concentrations of indolebutyric acid (40 to 60 mg./l., 20 mg./cc., and 5 to 12 mg./g.). In addition to the 11 varieties tested during 1938 and for which 50 to 100 per cent root-

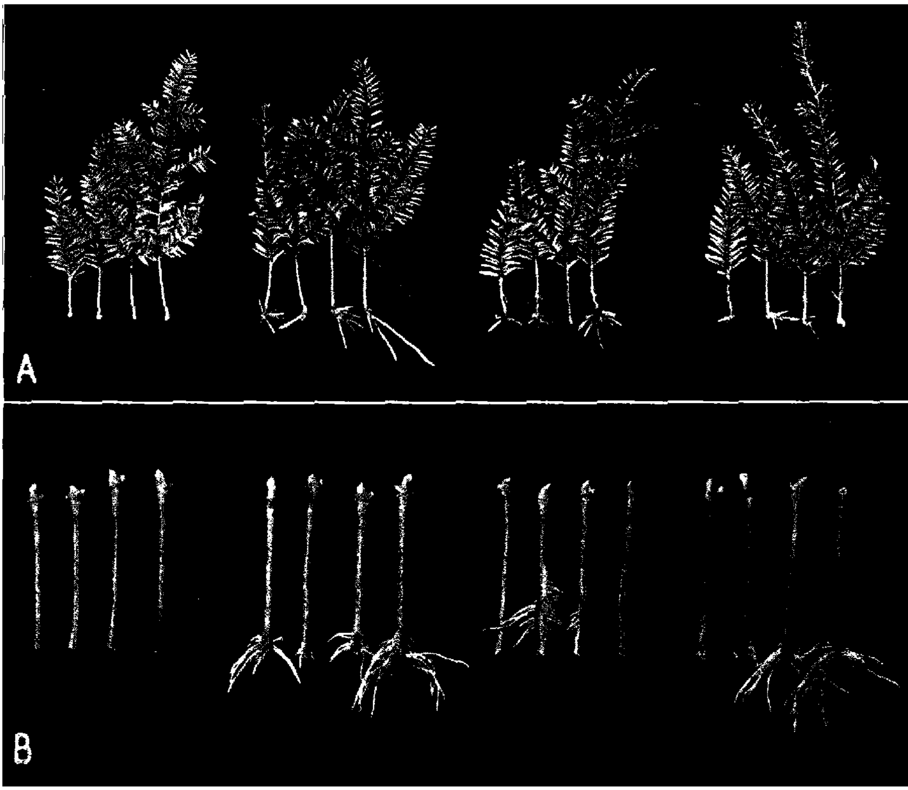

FIGURE 3. Comparative rooting for indolebutyric acid applied according to different methods to (A) *Tsuga canadensis* and (B) Concord grape. Left to right (A and B): non-treated control, 40 mg./l. (24 hours), 4 mg./cc. in 50 per cent alcohol (dip method), and 5 mg./g. powder (dip method).

ing was obtained in 40 to 60 days, two other varieties (Reine Elizabeth and Siebold) gave 100 per cent rooting in 25 to 28 days when treated with indolebutyric acid in a concentration of 12 mg./g. (Table IV). These results indicate that the named varieties of *Syringa vulgaris*, generally referred to as French lilacs, can be rooted readily and there should be no necessity in the future for grafting them. Rooted cuttings from the 1938 tests made an excellent growth in the field during the spring of 1939. Results with lilac are similar to those obtained with *Taxus cuspidata* and *Tsuga canadensis* for cuttings treated in June. Control cuttings of *T. canadensis* were previously rooted only when taken from June to August (2). However, seasons vary from year to year so that specific dates cannot be considered as an accurate indication of the relative age of the shoot.

TABLE IV

ROOTING OF LILAC (SYRINGA VULGARIS) TREATED WITH INDOLEBUTYRIC ACID APPLIED AS A POWDER. AVERAGE NUMBER OF ROOTS PER CUTTING

Variety	Day treated (May)	Root counts made (June)	Total No. days	Conc. of acid in mg. per gram talc			
				0*	2	5	12**
Arthur Wm. Paul	19	12	24	0	9	3	23
Clara Cochet	19	23	35	0	2	0	4
Perle von Teltow	19	23	35	0	0	0	9
Dame Blanche	15	23	39	0.3	1	1	7
Siebold	18	14	27	0	3	2	13
Toussaint Louverture	18	16	29	0	2	1	6
Reine Elizabeth	19	13	25	0	7	3	18
Reine Elizabeth	19	16	28	0	0.3	2	12
Prof. Sargent	19	16	28	0.3	4	4	21
Prof. Sargent	15	23	39	0	3	1	14
Mme. Florent Stepman	10	16	37	0	1	8	0
Mme. Florent Stepman	15	23	39	0	2	0	4
Column totals				0.6	34.3	26	131

* Finely ground control talc corresponding to that used in mixtures of the acid and talc.
** 100 per cent rooting obtained with this concentration except in the case of Mme. Florent Stepman.

Cuttings of many species of plants required higher concentrations of indolebutyric acid when taken during October and November as compared with other times of the year. The following species or varieties exhibited the differences mentioned: *Taxus cuspidata*, *Thuja occidentalis* varieties *globosa nana*, *robusta*, *spiralis*, and *ellwangeriana*, *Juniperus chinensis* var. *pfitzeriana*, *Chamaecyparis obtusa* varieties *filiformis* and *filifera aurea*, *Tsuga canadensis*, *Abies veitchii*, *Hibiscus syriacus*, Concord grape, *Ligustrum ovalifolium*, *catawbiense* hybrid rhododendrons, and *Celastrus articulatus* var. *punctatus*. Considering these results and others described in this section, it appears that regardless of the method of application a given concentration of a root-inducing substance does not produce optimum rooting for all species and not even for the same species at all times of the year and under all conditions normally met with in propagation procedures. In general, cuttings of evergreens have shown more variation than cuttings of deciduous plants. For this reason the approximate optimum concentrations for the two dip methods listed in Table I cover a wider range than in the case of evergreens treated according to the standard immersion method. Strict agreement in the results of different workers cannot be expected unless the principal factors which determine concentration requirements for a given root-inducing substance have been accounted for. The use of different root-inducing substances introduces additional factors which the results reported in other sections of this paper will show may be of a more limiting nature than those discussed in this section for indolebutyric acid.

TABLE V

COMPARATIVE RESULTS WITH ACIDS AND SALTS APPLIED BY DIFFERENT METHODS TO CUTTINGS OF EUONYMUS RADICANS. AVERAGE NUMBER OF ROOTS PER CUTTING

Treatment	Conc., mg. per unit of carrier	Indolebutyric		Naphthaleneacetic	
		Acid*	K-salt**	Acid*	K-salt**
Standard method 24-hr. treatment mg./l.	20	21	56	8	11
	0	1	—	—	—
Solution dip method mg./cc.	10	38	53	20	13
	4	51	44	10	6
	0	3	1	—	—
Powder dip method mg./g.	10	25	37	12	32
	4	14	31	8	11
	0	5	—	—	—
Column totals	—	158	228	58	73

* Dissolved in 50 per cent ethyl alcohol.
** Dissolved in water.

The effect of bottom heat (Table VI) shows that rooting of *Hibiscus* cuttings varied according to the temperatures used. Carnation responded in a manner similar to *Hibiscus*. In general, any beneficial effects of bottom heat (78° to 80° F.) consisted mainly of quicker rooting and a more rapid rate of root growth. Many species rooted better at 70° to 73° F. than at 78° to 80° F. These latter results were due primarily to the poor condition of the cuttings held at the higher temperature. No attempt was made in these tests to determine optimum temperatures for air and for the medium. The special case reported is used to illustrate the fact that a favorable temperature of the medium may act in effect the same as a higher concentration and hence temperature is a limiting factor when determining minimum concentration requirements for rooting of cuttings.

Optimum concentrations (75 to 100 per cent rooting) of indolebutyric acid powder for species not appearing in the tables are as follows: *Abelia grandiflora* Rehd. var. *rosea alba* (June 15) 2 mg./g.; *Chrysanthemum coccineum* Willd. (June 9) 2 mg./g.; *Cornus florida* L. (June 1) 5 to 12 mg./g.; *Corylus avellana* L. (June 16) 5 mg./g.; *Euphorbia pulcherrima* Willd. (May 1) 1 to 2 mg./g.; *Juniperus chinensis* L. var. *japonica* Vilm. (March 1) 5 mg./g.; *Lagerstroemia indica* L. (June 20) 2 mg./g.; *Lonicera japonica* Thunb. (April 14) 2 mg./g.; *Philadelphus pubescens* Loisel. var.

pendulifolius (May 16) 1 mg./g.; *Picea abies* Karst. (March 1) varieties *cupressina* 5 mg./g. and *echinaeformis* 2 mg./g.; *Prunus tomentosa* Thunb. (June 15) 2 mg./g.; *Pyrus serotina* Rehd. (June 1) 2 mg./g.; *Rhododendron mucronatum* Don. (May 31) 2 mg./g.; *Sophora viciifolia* Hance (June 15) 2 mg./g.; *Syringa vulgaris* L. varieties Mont Blanc (May 10) 2 mg./g., Oliver de Serres (May 18) and Ronsard (June 1) 12 mg./g.; *Tilia platyphyllos* Scop. (June 1) 2 mg./g.; *Vitex agnus-castus* L. (June 27) 5 mg./g. The date of treatment was specified in these tests (1939) since the time of year cuttings are taken is critical for some species, but the equivalent time may vary from year to year. For example, shoots of *Syringa vulgaris* varieties taken between the 10th and 15th of May, 1939, were comparable

TABLE VI

INFLUENCE OF BOTTOM HEAT ON ROOT FORMATION IN CUTTINGS OF HIBISCUS SYRIACUS TREATED* WITH INDOLEBUTYRIC ACID BY THREE METHODS. AVERAGE NUMBER OF ROOTS PER CUTTING

	Standard method, mg./l.					Powder dip method, mg./g.**					Solution dip method, mg./cc.			
	0	20	40	60	Group totals	0	6	18	35	Group totals	0	4	10	Group totals
Bottom heat 78° to 80° F.	0	4	4	11	19	3	16	26	53	98	0	19	34	53
Control bed 70° to 73° F.	0	2	3	6	11	2	7	5	1	15	0	6	9	15
Column totals	0	6	7	17	30	5	23	31	54	113	0	25	43	68

* Treated January 20 and data recorded 17 days later.
** Mixtures and control talc finely powdered.

in age and relative activity with those taken between April 28th and May 2nd, 1938. Shoot growth was correspondingly delayed on other species of plants during the spring of 1939.

Since control cuttings treated with talc showed consistently a slightly better rooting than non-treated controls or controls immersed in tap water, the effect of talc itself must be accounted for in experiments relating to results with comparative methods. In the case of young tip cuttings of chrysanthemum (Table II), talc controls remained in good condition whereas non-treated controls wilted each day for a period of several days and in some cases for the period of the test. Young carnation cuttings responded in a similar manner. This effect of talc appears to be due at least in part to differences which involve water relations. However, in many species there was no evidence of wilting but the talc controls showed better rooting and better callus formation than non-treated controls or tap water controls. The effectiveness of talc itself has also been observed by Stoutemyer (7). He reports percentage rooting values for cuttings treated with talc which represent in some cases much greater increases over the non-talc controls than any treatment with root-inducing substances over the talc controls (7, p. 818–819).

Results of some recent experiments indicate that the activity of control talc (Merck & Co. Inc.) may be due in part to the presence of a physiologically active ingredient which was obtained by extracting talc with chloroform. The chloroform residue was active when tested on tomato plants. This work is being continued for the purpose of determining the identity and the quantities of a physiologically active substance in different grades of talc and in other powders which might prove suitable as carriers for growth substances.

RELATIVE ACTIVITY OF DIFFERENT SUBSTANCES

There is general agreement in published reports as previously pointed out (4) that indolebutyric and naphthaleneacetic acids are more effective for rooting cuttings than indoleacetic acid, and that indolebutyric acid is the most effective of the three, from all standpoints, for practical use. Results with the concentrated solution and powder dip methods confirmed the results obtained with the standard immersion method of treatment in showing that indolebutyric acid was more effective on cuttings of most species than either naphthaleneacetic or indoleacetic acids. These experiments included the use of species requiring relatively low, medium, and high concentrations of the root-inducing substances (chrysanthemum, rose, *Euonymus*, *Hibiscus*, and California privet). Exceptions to the general rule just mentioned are known and have been reported (9, 4). For example, the fact that naphthaleneacetic acid is much more effective on privet than either indoleacetic or indolebutyric acids has been verified by tests in which the substances were applied as dilute solutions (5 to 80 mg./l.), as concentrated solutions (1 to 20 mg./cc.), as powders (1 to 12 mg./g.), and as vapors (10), the concentrations of which were not known. In a similar manner tests with all four methods showed that *Euonymus radicans* responded best to indolebutyric acid.

Whereas the optimum range of concentrations of indolebutyric acid is 1 to 80 mg./l. for the species listed in Table I, the optimum range of concentrations of indoleacetic acid for many of these same species is considerably higher as previously reported (4). Thimann and Delisle give 25 to 400 mg./l. as the optimum range of indoleacetic acid for all species which they tested and 100 to 400 mg./l. as the range for species difficult to root (8). Hubert *et al.* (5) found 100 to 150 mg./l. to be the optimum range for indoleacetic acid for species difficult to root. *Tsuga canadensis*, which is difficult to root (except during June and July when many controls have rooted), requires a concentration of indolebutyric acid of 40 to 60 mg./l. (Table I and Fig. 3 A). These results are to be contrasted with those reported by Thimann and Delisle, who found it necessarv to use indoleacetic acid in concentrations of 400 mg./l. to obtain equivalent (100 per cent) rooting of *Tsuga canadensis* cuttings (8). These results indicate that indolebutyric acid is about ten times more effective than indoleacetic acid for cuttings of *Tsuga canadensis.* Although cuttings of *Tsuga canadensis* taken during June root more readily than at other times of the year, they are difficult to maintain in good condition just as is described for the May shoots of lilac. In both cases the shoots had not attained their full length or leaf size. Treatment of cuttings with indolebutyric acid has not shown the age of the parent plant to be an important limiting factor, and certainly not the principal limiting factor, as claimed by Thimann and Delisle in the case of treatments with indoleacetic acid (8). Cuttings of *Tsuga canadensis* and *Picea pungens* taken from large trees (10 to 30 years old) rooted readily without the use of root-inducing substances when taken at the proper time of the year (2).

Potassium salts of indolebutyric, indoleacetic, and naphthaleneacetic acids were generally more effective than the acids as illustrated in Figure 2 H and Tables V and VII. The higher effectiveness of the salts for root formation was particularly noticeable in the case of the two dip methods. In most cases the tests constituted a comparison between the acid and salt of one or two substances. However, in one test the three principal acids and their corresponding potassium salts were compared simultaneously in concentrations of 2, 5, and 12 mg./g. on cuttings of chrysanthemum, rose, *Euonymus*, and privet. Although previously the salts were reported to be about equally as effective as the acids in the case of the standard immersion method of treatment (9, 4), the results obtained with the dip methods indicate that the salts are sufficiently more active than the acids to be of practical value in propagation. The potassium salts were more active at all three concentrations than the acids. In the case of the 12 mg./g. powder, the salt of naphthaleneacetic acid caused toxic and over-treatment effects on privet cuttings characteristic of an excessively high concentration. These effects consisted of excessive swelling and proliferation of the stems, the formation of many short roots which were inhibited in growth, and the distortion of the terminal growth. The newly formed leaves were small, savoyed, and exhibited downward rolling at the margins. Similar abnormalities and toxic effects were not caused by lower concentrations of the salt or by the same concentration (12 mg./g.) of the acid.

The high effectiveness of the concentrated solutions (1 to 20 mg./cc.), particularly for species relatively difficult to root, makes this method worthy of further consideration. Cuttings which responded favorably to the concentrated solutions of the acid and the K-salt of indolebutyric acid were rhododendron hybrids, *Tsuga canadensis*, *Abies veitchii*, *Ilex opaca*, *Ulmus americana*, *Picea glauca* var. *conica*, several varieties of *Thuja occidentalis* and *Chamaecyparis obtusa*, *Juniperus* sp., *Camellia japonica* varieties *alba plena* and *chandleri elegans*, varieties of *Syringa vulgaris*, and certain varieties of the commercial fruiting apple. The effectiveness of the potassium salt of indolebutyric acid on hybrid rhododendron cuttings is of particular interest and indicates that this and other salts might prove of considerable practical importance.

TABLE VII

RELATIVE ACTIVITY OF ACIDS AND SALTS FOR INDUCING ROOT FORMATION IN HARDWOOD LEAFLESS CUTTINGS OF HIBISCUS SYRIACUS*

Form of substance	Indolebutyric, mg./g. or mg./cc.					Naphthaleneacetic, mg./g. or mg./cc.				
	0	4	10	20	Totals	0	4	10	20	Totals
A. Powder dip method**										
Acid	0	5	9	—	14	0	2	5	—	7
K-salt	0	8	19	—	27	0	3	7	—	10
B. Solution dip method										
Acid	0	18	17	28	63	0	6	10	2	18
K-salt	0	22	21	42	85	0	3	4	15	22

* Treated November 11 and data recorded December 19, 1938.
** Control talc and mixtures not finely powdered.

FACTORS INFLUENCING SOLUBILITY AND PENETRATION

Treating dry cuttings with powders as recommended by Grace (1) proved relatively ineffective on most species. Wetting the basal one-half inch of the cuttings with water just before dipping into the powders proved much more effective (Fig. 2 G and Table VIII). Wetting the cuttings with

TABLE VIII

EFFECT OF WETTING CUTTINGS OF TAXUS CUSPIDATA BEFORE TREATMENT WITH TALC PREPARATIONS OF ROOT-INDUCING SUBSTANCES. AVERAGE NUMBER OF ROOTS PER CUTTING AFTER TWO MONTHS*

Condition of basal ends when treated	Indolebutyric acid in mg./g. talc**					Totals	Naphthaleneacetic acid in mg./g. talc					Totals
	0	1	4	10	50		0†	1	4	10	50	
Dry	3	1	2	1	1	8	1	1	4	4	6	16
Wet	4	3	4	8	17	36	1	7	9	21	27	65

* Treated October 19, 1938.
** Control talc and mixtures not finely powdered.
† Normal controls.

50 to 95 per cent ethyl alcohol proved more effective than wetting with water. Since the acids are more soluble in alcohol than in water, these results indicate that the presence of a solvent on the treated surface of the stem increases solubility of the crystalline root-inducing substance. It was

found that more of a given powder (about three times by weight) adhered to the wet cuttings than to the dry cuttings. However, the fact that salts were more effective than the acids, and that wetting the cuttings with alcohol was more effective than wetting with water, indicates that solubility and possibly penetration were more important than the additional amounts of powder adhering to the surface of the cutting. The higher activity of the salts as compared with the acids is at least partly explainable on the basis of the higher solubility of the salts in water.

The question of whether the powder treatments are effective because they supply the cuttings with just the proper quantities of the root-inducing substance over a period of several days or weeks, appears to be answered in the negative by results of tests in which the powder was washed off at various intervals after being applied. Data for hardwood leafless

TABLE IX

EFFECT ON ROOT FORMATION OF WASHING BASAL ENDS OF TREATED HIBISCUS CUTTINGS AT DIFFERENT INTERVALS AFTER THE TALC PREPARATION HAD BEEN APPLIED*

Treatment	Average number of roots						Average length of roots (mm.)				
	Non-treated controls	Treated, not washed	Time cuttings were washed (after)				Treated, not washed	Time cuttings were washed (after)			
			2 hrs.	1 day	3 days	7 days		2 hrs.	1 day	3 days	7 days
Washed	0	25	19	33	31	31	31	6	26	15	16
Washed, retreated	0	32	26	38	58	49	18	6	7	10	6

* 25 mg. indolebutyric acid per gram of talc (control talc and mixtures not finely powdered).

cuttings of *Hibiscus* (Table IX) show that if the powder remains on the cutting for as short a period as two hours there was sufficient penetration to induce much better rooting than on control cuttings. There was relatively little difference in the rooting of cuttings washed one, three, and seven days respectively after application of the powder. A similar series of cuttings retreated after washing off the powder (Table IX and Fig. 2 F) showed a slightly increased rooting response with respect to the number of roots induced, but in all of the retreated lots the roots were shorter than in the case of the lot not washed and not retreated. Control lots had not rooted in this time. These results indicate that the principal action of the powder occurs within a relatively short time and probably in many cases within the first 24 hours. With respect to number of roots and length of roots (Table IX), retreatment appeared to act in effect the same as a higher concentration. In these tests all of the cuttings were planted after receiving the initial treatment and then they were removed for retreatment and planted as originally. The process of removing and replanting the cuttings may have been a factor in causing some of the differences described.

Data in Table III indicate that there is considerable penetration of the root-inducing substance through the bark of Briarcliff rose cuttings since covering the bark with paraffin before treatment with the powders reduced the number of roots which emerged. These results are in agreement with those previously reported for solution treatments (4). In both tests effective concentrations caused roots to emerge from tissue where roots do not normally emerge in control cuttings, even from tissue located several inches above the region where the substance was originally applied. Results with root-inducing substances applied as dilute or concentrated solutions, as powders, and as vapors all show that in the case of species responding to treatment roots emerge from stem regions characteristic for the species and also from other regions, including all levels from base to tip, depending upon the species and the concentration of the substance.

DISCUSSION

Tests with root-inducing substances show that optimum rooting varies with the species, the age and relative activity of the shoot, the time of year treatment is administered, the kind and concentration of substance, and the method of applying the substance to the cutting. In the case of all methods the concentration of root-inducing substance varied with the species tested. These results are in agreement with those reported extensively in the literature with particular reference to solution treatments. However, relatively few reports deal with the use of powder preparations. In the present tests the results show that the use of powder preparations of root-inducing substances involves essentially the same factors which have proved limiting in the case of solution treatments. The principal differences were quantitative.

With respect to concentration requirements on a weight basis the concentrated solutions (1 to 20 mg./cc.) and powder preparations (1 to 12 mg./g.) were about equally effective for a given species, but they represent values 10 to 1,000 times higher than equivalent concentrations (1 to 80 mg./l.) used for the standard immersion method of treatment. While young shoots of woody and herbaceous plants generally responded more readily than older shoots to lower concentrations of root-inducing substances, it was observed that young shoots of some species (lilac, privet, Japanese maple, hemlock) tolerated and required relatively high concentrations for optimum rooting.

Lilac cuttings responded only slightly or not at all to treatment with 2 to 5 mg./g. but they responded exceptionally well when treated with 12 mg./g. Data for lilac show that a proportional increase in rooting did not occur as a result of an increase in concentration from 2 mg./g. to 5 mg./g. (Column totals, Table IV), which is the lower part of the effective range. This is to be contrasted with the marked increase occurring with the use of 12 mg./g. which represents the higher part of the effective range. Since these results were typical of most other species difficult to root (rhododendron hybrids, English varieties of carnation, and certain evergreens), it is important when determining concentration requirements to include the entire effective range of concentrations. As representative of species difficult to root, the varieties of *Syringa vulgaris* are well suited for root formation tests. Tests covering a period of three years show conclusively that the nine varieties reported in the present paper and seven other varieties reported by Kirkpatrick (6) can be rooted successfully (50 to 100 per cent) when treated with indolebutyric acid applied as a solution or a powder and thus need not be grafted as has been the common practice in the past. Rooted cuttings of these varieties have shown a vigorous growth when planted in the field.

The age of the shoot proved to be an important limiting factor in the case of all methods of treatment. Likewise, the time of year cuttings were treated was an important factor. Not all the differences are explainable solely on the basis of the degree of maturity or on the basis of the relative hardness of the stem. Whereas the lilac rooted readily only from the young shoots taken in May, the Concord grape, *Hibiscus*, and many evergreens rooted more readily before and after the months of October and November. At least in some species the degree of dormancy of the buds appears to influence rooting. The age of the parent plant was not an important limiting factor in tests with woody species since all plants were more than five years of age and in most cases more than ten years old.

Indolebutyric acid was more effective than naphthaleneacetic acid or indoleacetic acid on most species. In our tests this held true for the same species regardless of the method used for applying the root-inducing substance. The higher activity of the salts as compared to the acids, for the rooting of both softwood and hardwood cuttings, indicates that solubility and penetration are important factors regardless of how the root-inducing substance is applied. Grace (1) also found the K-salt more effective than the acid. Results relating to the use of different solvents, removal of the test powder (by washing) at various intervals up to several days after treatment, and coating the bark with paraffin all show that treatments which tend to increase solubility or penetration were most effective for rooting. Since the principal effect of the powder treatment occurred within 24 hours or less, adherence of the powder to the treated surface of the cutting appears not to be of any considerable importance after the first day. According to our results there should be no necessity for planting cuttings with greater care in the case of powder treatments than with other methods. Any powder which may be dislodged when the cuttings are planted appears to be of little importance if the proper concentration of an active root-inducing substance is applied initially.

Control cuttings treated with talc produced more roots per cutting and in a shorter time, and produced a greater quantity of callus on some species as compared with either non-treated controls or tap water controls. These differences were consistent but not pronounced in comparison with responses to treatment with talc preparations containing the root-inducing substance. At least part of the effect of talc appeared to involve water relations since the talc controls exhibited less evidence of wilting at all times and the bases of the cuttings showed less drying and discoloration as compared with non-treated or tap water controls. A similar effect in reducing wilting has been observed in tests in which the roots of tomato seedlings were dipped into colloidal clay (a finely ground clay containing phosphates and used commercially as a fertilizer) at the time of transplanting from flats. In addition to its influence on water relations, control talc was found to contain a physiologically active ingredient which was extracted with chloroform. The results with control talc indicate that it is of considerable importance in accounting for any beneficial effects of the carrier, particularly in experiments relating to the use of powder or dust preparations of root-inducing substances. Talc induced twice as many roots on chrysanthemum cuttings as compared with the number of roots on non-treated or tap water controls.

The higher activity of powder preparations which were furnished in a finely ground state as compared with similar preparations prepared by mixing commercial grades of talc and the root-inducing substances (Table I) is a point of considerable importance not only in experimental work but also in practice. Thus the concentrations specified for optimum rooting of a given species will depend not only upon the kind of active root-inducing substance, but also upon the physical characteristics of the powder. Until some standard type of powder is used, it seems likely that there will not be entire agreement in the results of different workers. While our results agree in the main with those of Stoutemyer (7) there are certain differences with respect to effective concentrations which may be due in part to the differences in the physical characteristic of the test powders.

SUMMARY

Treatment of cuttings with root-inducing substances applied as relatively dilute solutions (1 to 80 mg./l.), as concentrated solutions (1 to 20 mg./cc.), and as powders (0.5 to 50 mg./g.) produced essentially the same rooting response. Concentration requirements for optimum rooting varied according to the kind and form of substance, the kind of carrier or solvent, the species of plant, the age and relative activity of the shoot, the time of year treatment was administered, and the method of applying the substance to the cuttings.

The physical characteristics of the test powders appeared to account mainly for the differences in the effective range in concentration of 4 to 50

mg./g. for the coarser test powder and 0.5 to 12 mg./g. for the finer test powder. The concentrated solution (1 to 20 mg./cc.) and powder (0.5 to 12 mg./g.) dip methods were about equally effective on a weight basis but represent concentrations 10 to 1,000 times those producing equivalent rooting according to the standard solution method of treatment (1 to 80 mg./l.).

Potassium salts were consistently more effective than the acids (indoleacetic, indolebutyric, and naphthaleneacetic) which appeared to be due in part to solubility relations. Indolebutyric acid or the salt was more effective on most species than indoleacetic or naphthaleneacetic acids. The high activity of potassium indolebutyrate suggests that it may be of considerable practical importance.

Talc controls exhibited better rooting than non-treated controls or tap water controls. At least part of the beneficial effect of talc appeared to involve water relations since at all times the cuttings showed less evidence of lack of water than did the non-treated or tap water controls. An ingredient of control talc, soluble in chloroform, was found to be physiologically active when tested on tomato plants.

Treatment of some 70 species included types readily rooted, moderately difficult, and those definitely difficult to root. Varieties of *Syringa vulgaris* rooted in 24 to 39 days (50 to 100%) when treated with indolebutyric acid applied as a powder.

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