

## **The influence of electrical charge and indolebutyric acid on rooting of willow cuttings**

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### **Abstract**

Willow cuttings were treated by direct electrical current (DEC), with a negative or a positive electrode inserted inside them, and with the other being outside. Both directions of DEC flow between electrodes stimulated rooting as actively as growth stimulators. The mung bean or oat coleoptile straight growth test showed that the cuttings released some growth promoters into the surrounding water. However, the simultaneous release of some undetected rooting inhibitors was not excluded.

The experiment, shows the possibility of changing the chemical regulatory mechanisms of tissues by the use of weak, direct electrical current which causes, the migration of particular substance(s) into or from a definite tissue or organ.

The formation of root primordia and their growth is controlled metabolically by the level and location of growth substances in the tissues of the cutting. Growth substances can stimulate the process of root formation (Domański 1967; Domański et al. 1969; Krelle and Libbert 1969; Walter 1968). In most cases auxins play the main role in this process. Their activity is influenced by the level of inhibitors (Turetskaya et. al. 1966; Wain 1970), or rooting cofactors (Hess 1964).

The differences in concentration of these substances at root initiation sites could be the source of variation in the process of rooting (Chin-Ting-Yun, et al. 1969; Domański et al. 1969; Julliard 1964; Kefeli 1975; Kundu and Audus 1974). Compounds which stimulate or inhibit rooting may have different locations in the tissues, different mobility and definite electrical charge (Good 1974; Milborrow 1974; Nichols et al. 1970). Changes in concentrations of these compounds at a given site could be achieved

when direct electrical current (DEC) flowed through the cutting in a radial direction.

The influence of electrical current on the physiological processes of plants has been investigated in several works (Evans 1974, Gunnar et al. 1968; Zvara 1969). For instance Evans (1974), Milborrow (1974) and others have shown that the electrical current affects the permeability of cytoplasmic membranes. The change in permeability causes a change in the bioelectrical potential of tissues (Evans 1974; Gunnar et al. 1968) which is connected with the influx of ions.

The main aim of this work was to detect if DEC may influence rooting of willow cuttings.

#### MATERIAL AND METHODS

The experiments were carried out in the spring of 1974 and 1975, on hardwood cuttings of *Salix daphnoides* acutifolia which in comparison with other varieties of willow have poor rooting ability (Domański 1967).

Willow cuttings were collected at the end of March. They were 20 cm long and about 10 mm in diameter.

The root primordia are formed in the tissues external to the cambium. Therefore, the experiments were carried out with DEC flowing

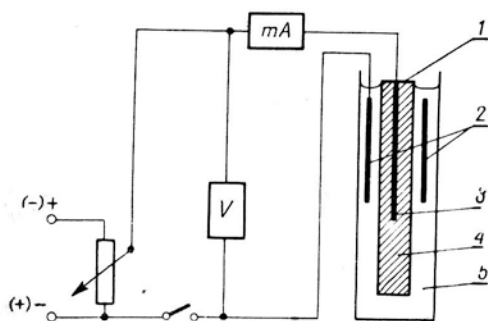


Fig. 1. The scheme of the electrical circuit

1 — basal end of the cutting, 2 — cross-section of the external electrode, 3 — electrode inside the cutting, 4 — the apical end of the cutting, 5 — distilled water

from the middle of the cutting to the outside of it, perpendicular to the surface of the cutting. A platinum electrode which was, 6 cm long, was introduced into the pith through the basal part of the cutting (Fig. 1).

The external electrode (also platinum 6 cm long) was shaped like a cylinder, which surrounded the cutting. The cutting together with both electrodes was immersed either in distilled water or in a solution of IBA 100 cm<sup>3</sup> in both cases. In some treatments, the IBA solution was used before the electrical treatment of the cutting in distilled water. The difference in electrical potential between the electrodes was regulated by the potentiometer, to obtain the value of DEC 3 mA. The DEC of this magnitude is usually applied for electrophoresis of organic acids. The value of the electrical charge was regulated during the time of the DEC action on the cutting. The experiments were carried out in two different ways: 1. the positive electrode was inside the cutting, or 2. the positive electrode was outside. The electrical charge of 0.1, 0.4 and 0.8 Coulombs, was given respectively for 30, 120 and 240 seconds. This treatment did not cause any visible injuries to the cuttings.

Table 1

The influence of electrical current on the rooting of willow cuttings  
(negative electrode was inserted inside the cuttings)

	Electrical charge in coulombs			
	0	0.1	0.4	0.8
Number of rooted cuttings	6.0a	15.0c	15.0c	10.0b
Number of roots per cutting	2.5a	4.0b	5.0bc	6.0c
Number of shoots per cutting	1.0	1.4	1.0	1.2
Length of shoots	3.0	3.5	3.1	2.6

LSD — value for the number of rooted cuttings at the 5% level = 3.1.

LSD — value for the number of roots per cutting at the 5% level = 1.4.

The cuttings which were treated by electrical charge, were then each placed in one tube with Konpp's solution. The tubes were put into the water bath with a temperature from 17°C (night) to 22°C (day). The cuttings were immersed in the water solution to 3/4 of their length. After 10 days, the observations were made on the number of rooted cuttings, and on the number of roots and shoots per cutting. The means, shown in the tables, are taken from three experiments each with 15 cuttings per treatment.

Bioassays were made to detect the kind and amount of growth regulators in the water in which the cuttings had been subjected to DEC.

Growth regulators were extracted with ether from the acidified water (pH 2.8-3.2). Bioassays were made using oat coleoptiles and the mung bean test for rooting substances. The absorption spectrum of the water, surrounding the DEC treated cuttings, was made with Beckman spectrophotometer in short waves of 290-400 nm.

## RESULTS

When the negative electrode was inserted inside the cutting and the positive outside it (Table 1), the stimulation of rooting was noticed. This stimulation was marked more at the lower electrical charges of 0.1 and 0.4 C. The charge of 8.0 C (coulomb) showed less stimulatory effect on the number of rooted cuttings but it increased markedly the number of roots per cutting. Stimulation of shoot formation was most evident at the low electrical charge of 0.1 C.

Table 2

The influence of electrical current and indolebutyric acid (IBA), on the rooting ability of willow cuttings (positive electrode was inserted inside the cutting);

	Electrical charge					
	0	0.4c	0.8c	0.8c + IBA * 200 mg/l	0.8c + IBA ** 200 mg/l	IBA 200 mg/l
Number of rooted cuttings	5.0a	11.0bc	14.0c	13.0c	14.0c	8.0ab
Number of roots per cutting	5.6a	5.8ab	6.3ab	7.3bc	8.2c	5.0a
Number of shoots per cutting	1.2	1.0	1.3	0.7	0.0	0.7
Length of shoots per cutting in cm	5.0	4.0	3.0	4.0	0.0	3.0

\* IBA was applied before the treatment of cuttings with electrical charge.

\*\* IBA was applied during this treatment.

LSD — value for the number of rooted cuttings at the 5% level = 3.4.

LSD — value for the number of roots per cutting at the 5% level = 1.6.

When the positive electrode was inserted inside the cutting (Table 2) the stimulation of rooting was also evident. The charge of 0.8 C was more effective than of 0.4 C in this case. Simultaneous action of IBA and of the electrical charge (Table 2) did not cause a substantial increase of the number of rooted cuttings and of the number of roots per cutting comparing with that received with electrical charge alone. It did not make much difference if IBA was used before or during the treatment with electrical charge (compare columns with asterix).

Shoot formation was completely inhibited when the electrical charge flowed through the cutting immersed in the IBA solution.

The water in which the cuttings were subjected to electrical charge ("+" inside) contained some compounds with spectra similar to that of phenols. As it is known, some inhibitors have phenolic nature. The compounds extracted from this water by ether, and tested by the bioassay of oat coleoptile segments caused inhibition of segment growth. This

indicates that some growth inhibitors were released from cuttings to the water when the electrical charge flowed from the "—" electrode being inside the cutting towards the "+" electrode being outside it. This was in agreement with the fact, that nearly all known inhibitors have a negative electrical charge. When the "+" electrode was inserted inside a cutting, the water surrounding it showed only slight inhibitory activity in oat bioassay.

The mung bean test showed that substances promoting root initiation were released to the external solution when the "—" or "+" electrode was inserted inside the cutting. More stimulation was found with this biotest when „—" electrode was inside the cutting.

### DISCUSSION

The explanation to the problem why removal of growth regulators from the cutting by application of DEC is accompanied by the stimulation of rooting is not easy. Possibly there are, besides growth promoters, some other substances which are also released to the water. Perhaps these substances although undetectable in most cases in mung or oat tests synergetically inhibit rooting when present in the tissues.

Rooting stimulation occurring when the positive electrode is inserted inside the cutting may be explained by the migration of rooting inhibitors from the bark and external layers of the xylem to the deeper layers of the xylem and to the pith, where they can not affect rooting.

From a general point of view, this experiment shows the possibility of using weak electrical current to cause the migration of particular substances to, or from a definite tissue (or organ) in order to change chemical regulation of its metabolism. This phenomenon may be compared to "electrophoresis *in vivo*". However, its nature is probably very complex and may involve several phenomena at the cell level as well as at the level of tissue or organ and the surface between the tissue and medium.

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## Wpływ ładunku elektrycznego i kwasu indolilomasłowego na zakorzenianie się sadzonek wierzby

### Streszczenie

Sadzonki *Salix daphnoides acutifolia*, o słabych zdolnościach ukorzeniania, były poddane działaniu prądu stałego (0,1; 0,4 i 0,8 kulomba), który płynął prostopadle do powierzchni w podstawowej części sadzonki. W czasie działania prądu, sadzonki były zanurzone w wodzie lub roztworze IBA.

Stwierdzono, że gdy elektroda ujemna była umieszczona wewnątrz sadzonki, wystąpiła widoczna stymulacja ukorzeniania przy 0,1 i 0,4 kulomba. Obserwowano też stymulację ukorzeniania, gdy dodatnia elektroda była wewnątrz sadzonki.

Traktowanie sadzonek roztworem IBA, przez który przepływał prąd stały (elektroda dodatnia w sadzonce), spowodowało zwiększenie się liczby korzeni na sadzonce ponad liczbę stwierdzoną u sadzonek kontrolnych, lecz jednocześnie zahamowało wzrost pędów.

Substancje uwolnione z sadzonki do roztworu na skutek działania prądu, wykazywały właściwości stymulatorów wzrostu, w teście ukorzeniania fasoli złotej.

Test koleoptyli owsa wykazał uwalnianie się inhibitorów wzrostu, gdy elektroda „+” była na zewnątrz. Nie jest jednak wykluczone, że jakieś inhibitory zakorzeniania mogły przejść do roztworu z sadzonki w przypadku, gdy elektroda „-” była na zewnątrz; nie zostały one jednak wykazane przez test koleoptyli owsa.

Doświadczenie to wykazuje możliwości powodowania zmian w biochemicznej regulacji metabolizmu tkanki lub organu poprzez zastosowanie słabego prądu stałego. Przepuszczając prąd przez dany organ można wprowadzić do jego tkanek pewne substancje jonowe lub też spowodować ich wydzielanie.