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Application of electric conductivity measurements to the estimation of frost injury in plant tissues*

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Electrical conductivity of, or resistance to flow of (weak) electric current through, plant tissue has long been used for estimating changes brought by various factors in the properties of semipermeable cell membranes (Ostrehout, 1918).

In normal growth conditions, tissue water is contained both inside and outside the living protoplast. The outside solution is distributed along cell walls, in intercellular spaces and in xylem vessels. It forms a continuous system favourable to diffusion of solutes. On the other hand, diffusion outwards the living protoplast is restricted by the semipermeable properties of cell membranes which form an effective barrier to diffusion. This effect is manifested in the resistance offered by living tissue to a weak current. According to Olien (1964), this resistance is as high as 5×10^5 ohms/cm along the longitudinal vein of a cereal leaf, when the weak direct current is applied. It falls below 5% of the initial value after the tissue has been killed; the semipermeable properties must have been destroyed, and the cell electrolites effused into intercellular spaces, whereby the electrical conductivity of the killed tissue was rapidly raised.

Changes in the electrical conductivity have been used for estimating tissue injuries caused by poisons, concentrated salt solutions, electric shock, and cold (Ostrehout, 1922; de Plater and Greenham, 1959; Wilner, 1961; Olien, 1961, 1964; Katz and Reinhold, 1964; Greenham *et al.*, 1966; Calder *et al.*, 1966).

The conductivity (or resistance) may be affected by such factors associated with the measuring technique as the type of electrodes used and their mutual distance, mode of attachment to plant tissue, potential applied, a.c. frequency, and properties of the tissue itself, e.g. water content. Data provided by the literature (Teske, 1965; Olien, 1964; Hayden *et al.*, 1969) are not consistent with each other what is probably due to the variety of studied plant tissue.

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It was the purpose of the present study to explore how the above factors affect resistance and to establish conditions providing for maximum reproducibility of data obtained on measuring frost injury in the tissue of cabbage and rape plants.

METHODS

Cabbage (*Brassica oleracea* L., cvar. Dittmar white) and winter rape (*Brassica napus* L., cvar Górczański) grown in a greenhouse were used for the investigations.

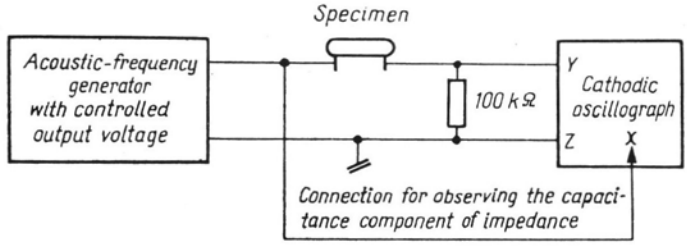

Fig. 1. Assembly for studying the effects of a.c. frequency and supply potential on leaf resistance.

Two platinum wire electrodes with pointed tips (0.05 mm in diam.) were driven into, but not through, the leaf. The effects of interelectrode distance, potential applied and a.c. frequency were studied in an assembly presented in figure 1.

In measurements of the resistance in relation to water content, the electrodes were held by a special claw which provided for a fixed penetration depth (0.5 mm) and constant pressure on the leaf surface and enabled the use of six electrodes at a time (Fig. 2). For the measurements a conductometer with a measuring range of 200 to 200,000 ohms was used.

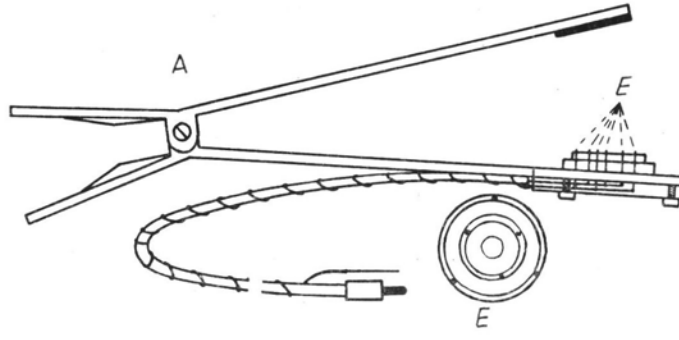

Fig. 2. Special device for inserting the electrodes into the leaf:
A — profile plan, E — disposal of electrodes (face plan).

The tissue was frozen in a SKP cold chamber at —5°, —7°, and —9°C for 2 hrs. The rates of freezing and defreezing were 7° and 8°C/min., respectively. Control, unfrozen leaves were kept at 5° for a period equal to that of freezing of other leaves. Water content was determined by drying the frozen and unfrozen leaves at 105°C.

RESULTS

Interelectrode distance

As evident from figure 3 when the interelectrode distance is kept below 4 mm the leaf resistance increases rapidly and within 1—4 mm the increment is about 70 kohms/mm. Above 4 mm the resistance rises less

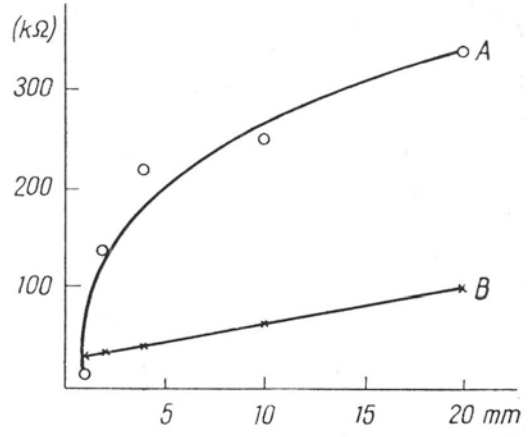

Fig. 3. Leaf resistance in relation to interelectrode distance:
A — in the parenchymatic tissue, B — along leaf vein.

rapidly, on the average to 8 kohms/mm. Therefore, the interelectrode distance should be kept at minimum 4 mm. The upper limit is chosen according to the type of material and the experimenter's covenience. In each case the distance must be established beforehand and kept constant.

Stabbing the leaf along the vein results in slight variation of the leaf resistance with the interelectrode distance, presumably because the resistance of cell sap is primarily measured.

Potential applied

As the potential is raised from 1 to 6 V, the leaf resistance is practically constant (Fig. 4). The range is therefore smaller than that suggested by K a t z and R e i n h o l d (1964) who give 7 V as the limiting value,

basing on Hörber's (1945) data, and much smaller than that applied by Teske (1965) who used potentials greater than 10 V.

In the present experiments, the resistance varied rapidly only after a potential of 10 V had been applied; this fact may be assocciated with both electrolytic polarization and current-effected changes in the semipermeable properties of cell membranes. The effect of the 10 V potential

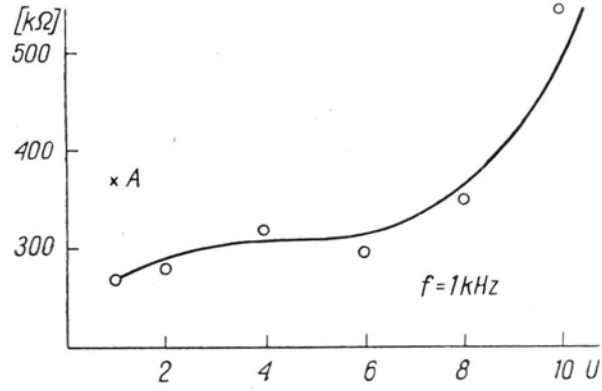

Fig. 4. Leaf resistance in relation to potential applied, (f = 1kH/s, interelectrode distance = 4.5 mm).
A — redetermination after application of 10 V.

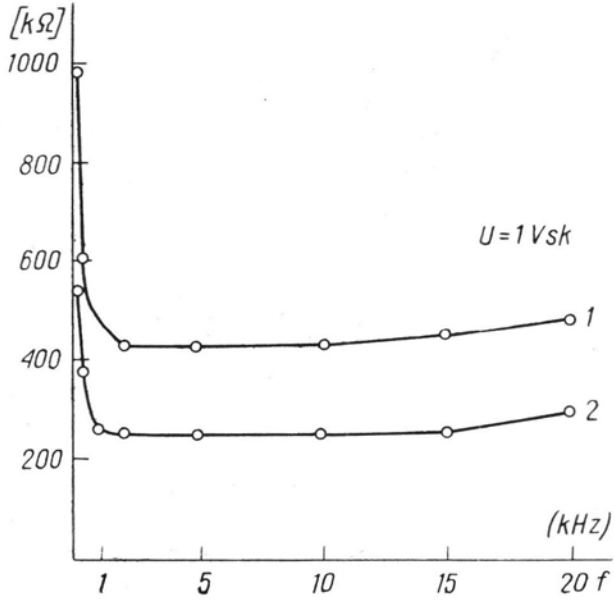

Fig. 5. Leaf resistance in relation to a.c. frequency, (interelectrode distance = 4.5 mm, potential applied = 1V, curves drawn for two different leaves).

on the permeability of cell membranes is shown by the fact that resistance measured at the point to which a potential of 10 V was applied was greater than that obtained prior to application of 10 V (Fig. 4). This fact points to flow and loss of some cell electrolite.

A. C. frequency

Application of direct current for measuring plant tissue resistance gives rise to interference by electrolytic polarization. Alternating current is therefore preferable. As evident from figure 5, the leaf resistance is practically unaffected by applied a.c. frequency, a fact consistent with theoretical expectation.

A rapid increase in the resistance observed at frequency lower than 1 kH/s is presumably due to polarization.

It was stated that the phase angle was 10° what means that the capacitance was caducous at the point of measurement.

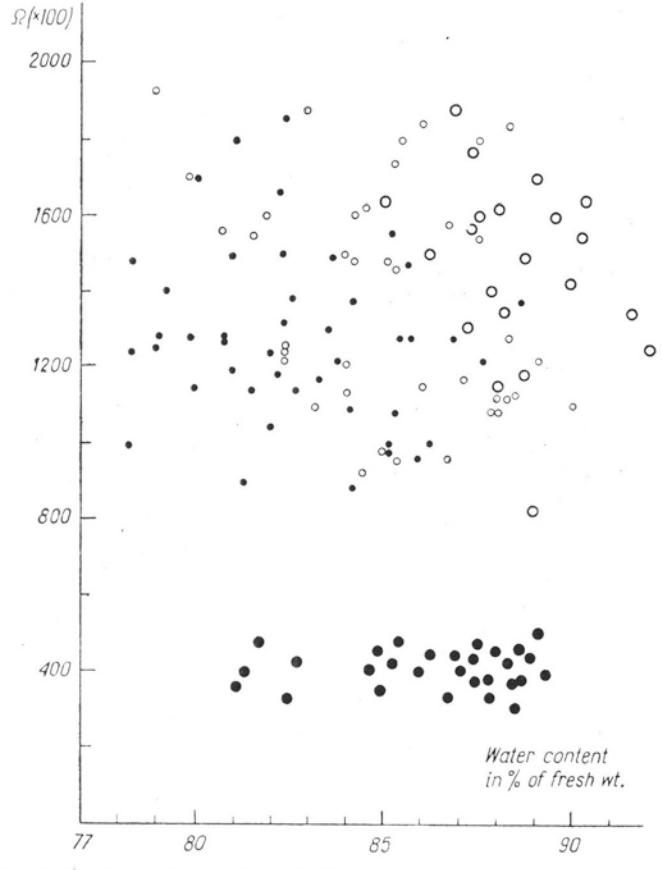

Fig. 6. Leaf resistance in relation to water content in leaves:
o — unfrozen leaf (young) O — unfrozen leaf (older) • — frozen leaf (surviving)
● — frozen leaf (killed)

Water content and viability of tissue

Resistance measured across various rape leaves is presented in figure 6. The leaf resistance appears to be practically unaffected by the over-all water content: the resistance of younger leaves with lower water content is much the same as that of the older ones. Low water content of 78—79% caused no distinct rise in resistance. On the other hand, the resistance fell rapidly in the leaves killed by freezing. Most of these leaves had a high water content (85—90%).

Interestingly, in the leaves that survived freezing the water content was considerably reduced, whereas the killed leaves contained nearly as much water as before freezing. The illustrative data are:

unfrozen leaves, av. 87.04% H_2O;
live frozen leaves, av. 82.11% H_2O;
killed frozen leaves, av. 85.79% H_2O.

This fact emphasizes the significance of the tissue's ability to transpire water rapidly on freezing for its frost hardiness.

CONCLUSIONS

The presented data allow the following conclusions:

1) In electrical conductivity measurements the interelectrode distance should be minimum 4 mm and the maximum potential applied 6 V. Frequencies higher than 1 kH/s have practically no effect on the value measured.

2) The overall water content in live leaf does not affect the value measured. But the permeability of cytoplasmic membranes to intracellular electrolites has a great effect on the resistance measured. With the cytoplasmic cell barrier destroyed by killing the protoplast, the resistance falls rapidly. With rape leaves this fall is about 70% of the initial value, i.e. somewhat less than that found by O l i e n (1964) for cereal leaves (95%).

3. Measuring the electrical conductivity of plant tissue allows to recognise rapidly irreversible frost injury at the point where it occurs.

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Zastosowanie pomiarów przewodnictwa elektrycznego tkanki do oceny uszkodzeń mrozowych roślin

Streszczenie

Praca miała na celu stwierdzenie, w jakim stopniu czynniki związane z techniką pomiaru przewodnictwa elektrycznego tkanki roślinnej, a także właściwości samej tkanki, wpływają na mierzone wartości oporu. Ustalono, że należy stosować nie mniejszy niż 4 mm rozstaw elektrod, a wartość przyłożonego napięcia nie może przekraczać 6 V. Częstotliwość napięcia powyżej 1 kHz praktycznie nie ma wpływu na mierzoną wielkość.

Ogólna zawartość wody w liściu nie miała większego znaczenia dla otrzymywanych wartości oporu, natomiast duży wpływ miała przepuszczalność błon cytoplazmatycznych dla elektrolitów wewnątrzkomórkowych. Zniszczenie mrozem cytoplazmatycznej bariery komórkowej powodowało duży spadek oporności.

Metoda pomiaru przewodnictwa elektrycznego tkanki (lub jej oporności) daje możliwość szybkiego rozpoznania nieodwracalnego uszkodzenia tkanki przez mróz, w miejscu powstania tego uszkodzenia.