tion the composition of the artificial rain water did significantly influence some changes in leaf Ca. At each concentration of Ca in the rain water, pH did not significantly affect the change in leaf Ca. At each level of pH, Ca in the rain water did not significantly affect the change in leaf Ca. However, the changes in leaf Ca during treatment with low pH, low Ca rain water were significantly different from changes in leaf Ca during treatment with high pH, high Ca rain (Table 3).

The effect of rain treatment on loss of leaf Ca was greater in the early summer than in the autumn period of observation. In the early summer period the young leaves were rapidly increasing in dry weight, but they were almost fully expanded. Tukey (6) cited several studies which reported an increased susceptibility to leaching of nutrients with increased leaf age. Addition of Ca to the artificial rain water in this study may explain the conflicting results. Not only were the reductions in leaf Ca less in the autumn period of observation, but there were significant increases in leaf Ca during the 7-hr treatments with high pH, high Ca rain water.

This study does not appear to confirm previous reports (1, 7) that in comparison with high pH, low pH rain leached more Ca from leaves. However, there were 2 important differences between this and the previous studies: spruce (species not given) (1) and maple (*Acer saccharum* Marsh) (7) trees were used in the previous studies; leaf Ca was measured in this study, whereas leached Ca was measured in the previous studies. If Ca moved rapidly into the leaves during the treatment period, low pH rain could increase Ca leaching without significantly affecting the level of Ca in the leaves.

Data collected in this study suggest that the recent apparent decline of leaf Ca in New York apple leaf samples (2) was probably not caused by acid rain.

Literature Cited

1. Abrahamsen, G., B. Kristian, R. Hornveldt, and B. Tveite. 1976. Effects of acid precipitation on coniferous forest. Research Rpt 6/76, Impact of acid precipitation on forest and freshwater eco-systems in Norway. The SNSF-project of the Agric. Res. Council of Norway.
2. Blanpied, G. D. and G. H. Oberly. 1978. Calcium and magnesium levels in the annual rings of 'McIntosh' apple wood. *J. Amer. Soc. Hort. Sci.* 103:638-640.
3. Cogbill, C. V. and G. E. Likens. 1974. Acid precipitation in the Northeastern United States. *Water Resources Res.* 10:113-1137.
4. Dittenhoefer, A. C. and B. E. Dethier. 1976. The precipitation chemistry of western New York State: a meteorological interpretation. Res. Proj. Tech. Completion Rept. for Off. Water Res. & Tech. (U.S. Dept. Int.) Proj. No. A-044-NY; 1-45.
5. Likens, G. E. 1972. The Chemistry of Precipitation in the Central Finger Lakes Region. Cornell University Water Resources & Marine Sci. Center Tech. Rpt. 50.
6. Tukey, H. B., Jr. 1970. The leaching of substances from plants. *Annu. Rev. Plant Physiol.* 21:305-324.
7. Wood, T. and F. H. Bormann. 1975. Increases in foliage leaching caused by acidification of an artificial mist. *Ambio* 4:169-171.

HortScience 14(6):708–709. 1979

Relationship between Cold Hardiness and Normalized Electrical Impedance in Unfrozen 'Delicious' Apple Shoots[1]

D. R. Evert[2] and D. R. Cooley[3]

Department of Plant and Soil Science, University of Vermont, Burlington, VT 05405

Additional index words. Malus domestica, cold injury, shoot diameter

Abstract. **Normalized electrical impedance and cold hardiness were measured for internodal sections of 'Delicious' apple (*Malus domestica* Borkh.) during fall and winter for 2 years. Each year, the normalized impedance for the August date was significantly higher than on later dates. The relationship between normalized impedance and cold hardiness was inconsistent; r^2=0.03 and P>5%, first year and r^2=0.4 and P<1%, second year. The relationship between cold hardiness and the mean temperature for the 7 days before the sample date was consistent; r^2=0.85 and P<1%, first year and r^2=0.81 and P<1%, second year. The slope of the line relating sample diameter and normalized impedance changed from plus to minus each year.**

Electrical measurements have been used to evaluate cold injury in woody tissues (1, 2, 4, 6, 9). Some methods predicted cold hardiness directly, without the need for laboratory freezing (15, 16), but variability prevented a precise estimate of hardiness. We determined the relationship between normalized impedance and cold hardiness of field collected shoots of 'Delicious' apple, and therefore, the possibility of using normalized impedance as a predictor of hardiness during the fall and winter.

Current year growth of seven 24-year-old 'Delicious' apple trees on Malling (M) 7 interstems and Robusta 5 rootstocks at the University of Vermont Horticultural Research Center, Burlington, were sampled on several dates during the fall and winter of 1974-75 and 1975 (Table 1) as previously described (2). Internodal sections were cut from the shoots and pooled. Ten to 23 sections, depending on the date, were selected randomly for electrical impedance measurements at 50 Hz and 500 kHz without laboratory freezing. Normalized impedance (Zn) was calculated from the impedance measurements by the formula: Zn= (Z50Hz − Z500kHz)/Z500kHz where Z50Hz and Z500kHz are the impedance magnitudes at 50 Hz and 500 kHz, respectively. Diameter measurements to the nearest 0.01mm were made after the impedance measurements.

Eight to 12 samples per test temperature were selected randomly from pooled sections. Following controlled freezing at rates of 5°C/hr or less (2), the samples were incubated for 2 weeks at 30°, split lengthwise, and rated on the following scale: 1 = no browning or slight browning at the ends; 2 = some browning throughout; 3 = complete browning. The temperature needed to produce a mean visual rating of 1.5 was the estimated cold hardiness and was computed using linear interpolation between the 2 temperatures with mean visual ratings on either side of 1.5.

The mean normalized impedance on August dates was significantly larger than on remaining dates (Table 1). Normalized impedance apparently predicted the start of the first stage of cold hardiness which has been associated with a translocatable factor produced in response to short days (5, 7) and

[1]Received for publication May 2, 1979. Vermont Agricultural Experiment Station Journal Article No. 421. We thank Dr. N. E. Pellett for his assistance in the preparation of the manuscript. This study was supported in part by a grant from the Horticultural Research Institute, Washington, D. C.

The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper must therefore be hereby marked *advertisement* solely to indicate this fact.

[2]Present address: Department of Horticulture, Coastal Plain Experiment Station, Tifton, GA 31794.

[3]Present address: Department of Plant Pathology, Fernold Hall, University of Massachusetts, Amherst, MA 01002.

Table 1. Properties of internodal sections of 'Delicious' apple as a function of date.

Date sampled	Mean normalized impedance[z]	Mean temp (°C)[y]	Estimated hardiness (°C)	Normalized impedance vs. diameter in mm
1974				
Aug. 19	2.76 a	--	--	0.17 D + 2.06
Sept. 5	1.91 b	--	--	0.20 D + 1.09
12	1.43 c	19.8	−6.2	0.07 D + 1.11
19	1.76 b,c	19.1	−7.5	0.11 D + 1.18**
24	1.42 c	--	--	0.04 D + 1.24
Nov. 6	1.40 c	--	--	0.03 D + 1.26
21	1.37 c	--	--	−0.03 D + 1.17
22	1.95 b	−1.0	−24.5	0.15 D + 1.17
1975				
Jan. 17	1.69 b,c	1.3	−32.6	0.05 D + 1.48
Feb. 1	1.54 b,c	−3.9	−30.7	−0.05 D + 1.76
13	1.88 b	--	--	−0.23 D + 3.02**
28	1.87 b	--	--	−1.33 D + 7.22
March 24	1.86 b	0.4	−20.6	−0.02 D + 1.97
April 11	1.75 b,c	--	--	−0.01 D + 1.82
Aug. 23	2.35 a	20.3	−6.3	0.02 D + 2.24
Sept. 4	2.05 b	16.1	−7.5	0.08 D + 1.73
17	1.81 b,c	12.3	−8.0	0.16 D + 0.97**
Oct. 1	1.66 c,d	13.5	−12.5	0.02 D + 1.51
15	1.79 b,c	8.2	−17.3	−1.75 D + 7.40
29	1.44 d	--	--	−0.04 D + 1.61
Nov. 12	1.55 c,d	--	--	--
26	1.46 d	1.6	−23.0	−0.05 D + 1.65
Dec. 10	1.65 c,d	−3.5	−27.3	−0.02 D + 1.72
23	1.38 d	−11.9	−23.8	−0.16 D + 2.06**

[z]Mean separation within each fall-winter by Scheffe's multiple comparison test, 5% level.

[y]Mean temperature for 7 days preceding date sampled.

**Correlation coefficients significant at the 1% level.

vegetative maturity (12, 13). Electrical impedance measurements are sensitive to membrane changes (3, 9), and we anticipated a decrease in normalized impedance as the plants became cold hardy.

A decrease in normalized impedance would confirm the increase in membrane permeability that was reported to occur during cold hardening and that paralleled seasonal changes in cold hardiness (8).

Our data indicated little or no relationship between the second stage of cold acclimation and normalized electrical impedance (Table 1). From September 12, 1974 to January 1, 1975, the estimated hardiness went from −6.2 to −32.6°C and the normalized impedance increased from 1.43 to 1.69 (Table 1). Similarly, from October 1, 1975 to December 10, 1975, estimated hardiness went from −12.5 to −27.3°C with essentially no change in normalized impedance. Estimated hardiness and normalized impedance for the dates where both quantities were measured showed a correlation only in the fall-winter period of 1975 when r^2=0.4 and P<1%; in 1974-75, r^2=0.03 and P>5%. These data indicate normalized impedance of unfrozen samples was not an acceptable predictor of cold hardiness. By contrast, estimated hardiness and mean temperatures for the 7 days prior to date of sampling were well correlated each year; in fall-winter 74-75, r^2-0.85 and in fall-winter 75, r^2=0.81 with P<1% each year.

The failure of normalized impedance to consistently correlate with cold hardiness indicated no pronounced increase in membrane permeability. Our results agree with those of McKenzie et al. (11) that membrane permeability increased only during the earliest stages of cold acclimation.

We observed a highly significant correlation between normalized impedance and diameter on two dates each fall and winter (Table 1). Also, the slope of the relationship of normalized impedance and diameter changed from plus to minus each winter (Table 1). The probability of such a change was calculated using the Mann-Whitney test (10): for fall-winter 74-75, P<0.2% and for fall-winter 75, P<1.6%. These changes in sign may reflect the differential change in seasonal hardiness of bark and wood described by Parker (14). Other research has established that bark and wood of plants have different normalized impedance values (3). Further research is needed to determine the cause of the change in sign of the slope of the normalized impedance vs. diameter equation.

Literature Cited

1. Blazich, F. A., D. R. Evert, and D. E. Bee. 1974. Comparison of methods of measuring winter hardiness of internodal stem sections of *Forsythia intermedia* 'Lynwood'. *J. Amer. Soc. Hort. Sci.* 99:211-214.
2. Cooley, D. R. and D. R. Evert. 1979. Normalized electrical impedance evaluates injury to stem sections of 'Delicious' apple stems. *J. Amer. Soc. Hort. Sci.* 104:561-563.
3. Evert, D. R. 1973. Factors affecting electrical impedance of internodal stem sections. *Plant Physiol.* 51:478-480.
4. ________ and C. J. Weiser. 1971. Relationship of electrical conductance at two frequencies to cold injury and acclimation in *Cornus stolonifera* Michx. *Plant Physiol.* 47:204-208.
5. Fuchigami, L. H., D. R. Evert, and C. J. Weiser. 1971. A translocatable hardiness promoter. *Plant Physiol.* 54:29-35.
6. Glerum, C. 1970. Vitality determinations of tree tissue with kilocycle and megacycle electrical impedance. *For. Chron.* 46:63-64.
7. Howell, G. S. and C. J. Weiser. 1970. The environmental control of cold acclimation in apple. *Plant Physiol.* 45:390-394.
8. Levitt, J. and G. W. Scarth. 1936. Frost-hardening studies with living cells. *Canadian J. Research* C14:285-305.
9. Luyet, B. J. 1932. Variation of the electrical resistance of plant tissues for alternating currents of different frequencies during death. *J. Gen. Physiol.* 15:283-287.
10. Meddis, R. 1975. A statistical handbook for non-statisticians. McGraw-Hill, London.
11. McKenzie, J. S., C. J. Weiser, E. J. Stadelmann, and M. J. Burke. 1974. Water permeability and cold hardiness of cortex cells in *Cornus stolonifera* Michx., a preliminary report. *Plant Physiol.* 54:173-176.
12. Nissila, P. C. and L. H. Fuchigami. 1978. Xylem water potential and electrical impedance ratios as measures of vegetative maturity in red-osier dogwood (*Cornus stolonifera* Michx.). *J. Amer. Soc. Hort. Sci.* 103:708-709.
13. ________ and ________. 1978. The relationship between vegetative maturity and the first stage of cold acclimation. *J. Amer. Soc. Hort. Sci.* 103:710-711.
14. Parker, J. 1962. Seasonal changes in cold resistance and free sugars of some hardwood tree barks. *Forest Sci.* 8:255-262.
15. Van den Driessche, R. 1973. Prediction of frost hardiness in Douglas fir seedlings by measuring electrical impedance in stems at different frequencies. *Can. J. For. Res.* 3:256-264.
16. Wilner, J. 1967. Changes in electrical resistance of living and injured apple shoots during winter and spring. *Can. J. Plant Sci.* 47:469-475.