A COMPARISON OF METHODS FOR THE DETERMINATION OF LOW TEMPERATURE INJURY TO PEACH TREES

by

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INTRODUCTION

The ability of plants to withstand low temperature can be called winter hardiness. Most annual plants escape such adverse conditions during their life period, but perennial plants must be adaptable to varying ecological conditions. Much early work on hardiness was unsuccessful because the knowledge of the nature of winter injury and the factors involved therein was lacking. According to Asai (1) low temperature injury occurs mainly due to freezing of tissues.

The temperature of the inside of a tree trunk was first measured by Hunter 1779 according to Levitt (16). During the beginning of the 20th century winter hardiness of plants was determined by field survival counts as described by Levitt (16). In 1927 Akerman treated woody perennial plants in freezing chambers at known temperatures and shifted them to greenhouses where the percentage of injury was estimated. He also standardized the conditions for artificial freezing. In the years 1930-32 Dexter et al (7) measured plant hardiness by the electrical conductivity method and concluded that the electrical current conducted was proportional to the injury observed.

Many plant hardiness investigations were made to determine at what temperature freezing injury occurs in different species and varieties of plants. This information was utilized in breeding for greater winter hardiness and for determining varietal adaptation. Two electrical methods have been used to measure low temperature injury--the exosmosis method as explained by Dexter et al (7) and the direct method of measurement as explained by Filinger and Cardwell (11).

Dexter (7) was the first man to use the exosmosis method. The measure of the electrical conductivity of the water solution in which cut sections of plant tissue were placed for a given period, gave an indication of the degree of injury. The diffusion of electrolytes was more rapid from damaged than from undamaged tissue. It was concluded that the higher the conductivity the greater the injury.

Filinger and Cardwell (11) used a rapid electrical method to determine plant injury by low temperature. They concluded that when the plants were killed or injured by freezing temperatures they offered less resistance to electrical currents as compared to living tissues. The advantage of this method was explained by them as being a simple, accurate and rapid method to measure injury without destroying the plant tissues.

According to Tysdal (22) hardening of plants before subjecting them to freezing temperature reduces injury. The different factors that influence hardening are soil moisture, light and temperature. Asai (1) suggested that the physical property of the protoplasm appears to be a prime factor involved in winter hardiness.

The object of the present investigation was to determine whether there was positive correlation between the direct reading method and the exosmosis method of measuring relative hardiness of plants subjected to different low temperature treatments. The relationship between twig diameter and low temperature injury was studied. An effort was made to determine if the distance between the electrodes affected the direct method of injury measurement.

REVIEW OF LITERATURE

Asai (1) reported certain facts on low temperature injury from his research on garden roses. Ice formation in intercellular spaces was found to be the cause of freezing injury. Rapid freezing caused more injury than gradual freezing. Temperature drop at the rate of 16° F. per hour caused more injury than a drop at the rate of 1° F. per hour. Rapid thawing also caused a high degree of injury. Thawing of frozen canes in water at 0° C. was nine times as rapid as thawing in air at 20° C. According to him, the lower the moisture percentage of the canes the greater the hardiness and vice versa.

Tysdal (22) worked with plants of different alfalfa varieties in an effort to determine the optimum temperature, light and soil moisture requirements for hardening. He determined that temperatures of 10-12° C. were best for hardening. Plant survival was increased with a decrease in the duration of light from 10.5 to 7 hours while hardening. Short days hardened the plants more than long days with other conditions being constant. The variation in the soil moisture requirement for hardening was so negligible he could not compute the result in all varieties; however, high soil moisture during freezing was found to increase survival. According to him the low temperature hardiness response of short day plants was due to an increase in soluble nitrogen or amino acids. Plants hardened under white light showed more hardiness as compared to those under red or blue light. To determine the duration of hardening, he treated elm trees from 1 to 35 days at 2-4° C. Fourteen to 15 day treatments were found to produce maximum hardiness. Hardiness remained constant from 15 to 23 days; beyond 23 days a decrease in hardiness was noticed.

Harvey (15) found 0° C. for five days to be the best temperature for hardening elm trees.

Dorsey and Bushnell(8) found that horticultural practices had an influence in developing hardiness in plants. Certain root stocks caused early maturity of wood of the scion variety. Late maturity of shoots due to heavy pruning has also been noticed. Peach trees injured by low temperature recovered much earlier when fertilized during early spring with about four pounds nitrate of soda than nonfertilized trees. Irrigation in early winter appeared to protect the trees from winter injury. Hardening of the plants prevented protein precipitation during freezing. According to these authors, injury occurs mainly due to the diffusion of water through the cell walls into the intercellular spaces resulting in the cells drying out. Also, they noticed varieties that were early maturing and had long rest periods possessed greater hardiness than late maturing ones.

From a large collection of data Max Pfaender (20) came to the conclusion that closely spaced, early planted apple trees survived winter injury better than those spaced farther apart and planted late. Mulched orchard trees showed considerable more winter hardiness than unmulched trees.

According to Dexter (6) a high available carbohydrate supply and controlled vegetative growth before cold weather induced hardiness to a maximum extent. Therefore carbohydrate synthesis should be proportionately higher than respiration while hardening plants. During hardening, light increases the photosynthetic products at the same time low temperature reduces respiration resulting in a low rate of carbohydrate degradation. When CO_2 was restricted in the hardening chamber, low temperature hardiness dropped greatly. He measured low temperature hardiness by the exosmosis method.

Because of thinness of the periderm, as well as relatively more active growth, roots are damaged from low temperatures more than shoots according to Craig (5).

When low temperature injury was observed by Greenham and Daday (14), they found that different locations (diameter) of woody plants had little effect on injury. According to them, the amount of injury was the same throughout the plant when exposed to freezing temperature.

Dexter et al (7) worked with alfalfa roots and measured the injury by electrical conductivity measurements which were supplemented by calorimetric tests for chlorides. Plant tissues were found to change their physical and chemical properties as a result of freezing temperature; therefore exosmosis of electrolytes into the external solution occurred. The diffused electrolytes were estimated by electrical conductivity measurements. Roots from three different alfalfa varieties were washed and dried, after which 20 gram samples from each of the varieties were cut into half-inch pieces and transferred into test tubes, stoppered, and frozen for about four hours at -8 to -9° C. After freezing, samples from each variety were planted in a greenhouse to observe their recovery. Fifty ml. of distilled water was added to the remaining test tubes and kept for ten hours at 25° C. The electrical conductivity of the extract was measured with a conductivity bridge. They noticed considerable varietal variability with respect to injury. According to them the higher the electrical conductivity the greater the injury.

Using the same experimental procedure as Dexter, Swingle (21) also investigated low temperature hardiness by the exosmosis method. He used root samples of different varieties of apples and found after subjecting them to low temperature that the smaller the diameter the greater the proportion of living cells, hence greater resultant injury at a given low temperature. He used a radio type conductivity meter with platinum electrode needles.

According to Merrill (18) exosmosis was not a causal injury but simply an incidental effect of low temperature and also an indirect relation to the inimical condition of the plant part placed in distilled water. He did emphasize that the exosmosis method cannot be a very accurate method to measure winter injury.

Wilner (26) in his apple varietal experiment of winter hardiness observed all the varieties showed negligible differences in their type of growth when measured by electrical conductivity method during the active growing season of July 1958. The electrical conductance ranged from 800 to 1350 micromhos. However, the conductance of diffused electrolytes declined rapidly as twig maturity advanced. When he treated the plants at different low temperatures, varieties showed variable electrical conductance and survival. The Antonovka apple variety with 71 per cent survival measured 296 micromhos on an average; McIntosh, 53 per cent survival, 445 micromhos; Northern Spy, 28 per cent survival, 661 micromhos. According to him, the higher electrical conductivity from hardened twigs generally signified greater percentage of twig killing. When the temperature was lowered from 0° F. to -10° F., a higher value of electrical conductivity was observed in all varieties with a resulting decrease in percentage of survival.

Filinger and Cardwell (11), using a direct reading method, observed that when plants were injured by extremes of temperature, they offer less electrical resistance than when alive. They measured low temperature injury using a 10 inch distance between electrodes and found 72 to 82 per cent drop in electrical resistance.

Campbell and Ghosheh (2) measured low temperature injury of selected grape varieties by the direct reading method and found a varietal response for injury resulting from different temperature treatments. A distance of 6 inches between the electrode needles was used to measure possible injury to grape canes treated under temperatures of -10° F., -17° F., -25° F. and -40° F. for 24 hours. Comparing the readings of the cuttings with those of rooted plants, they found relatively low hardiness in rooted plants likely due to vigorous vegetative growth.

According to Campbell (3) critical low temperature in the peach tree varies greatly with varieties and also between individual plants within a variety. Generally freezing injury causes blackheart of the branches.

Wilner (27) preferred laboratory tests for winter hardiness by electrolytic methods to save space and labor. His object of study was to determine the consistancy and reliability of electrolytic techniques. He found significant differences between varieties and between treatment temperatures. Certain direct relationships between tissue injury and amount of exosmosis solutions were reported by him.

Wilner (28) exposed some apple trees to outdoor winter temperatures and measured the injury by the direct reading method. He observed that shoots and roots offer different amounts of electrical resistance. From the electrical resistance measurements he predicted the field survival. He correlated the two methods of testing on indoor and outdoor treated plants. According to him both methods of testing gave reliable results, but the direct reading method gave a higher degree of accuracy than the exosmosis method.

MATERIALS AND METHODS

Preparation of Plant Material

Dormant one-year old, mostly single whip Golden Jubilee peach trees were used as experimental material. Irregular and injured roots were pruned from the trees and planted in gallon cans containing a soil mixture of 3:1 sandy loam and peat moss respectively. Altogether about 70 plants were planted and the best 60 selected for the experimental study. Immediately after planting the cans were watered thoroughly. No further watering was done during the hardening process. All the planted cans were removed to a thermostatically controlled cold storage room for hardening.

Hardening Process

The temperature of the cold storage room was maintained at 38° (±3°) F. throughout the period of hardening. White fluorescent lights with an average intensity of 32.8 watts per square foot were turned on for about 10 hours each day. Plants were hardened all at one time for about a 30 day period.

Freezing

Batches of fifteen plants were selected each time and the cans were wrapped with thin aluminum sheets covered with a fibrous insulating material. Different batches of plants were treated at three temperatures, -20, -10 and 8° F. separately in the freezing chamber. The insulation protected the root system from severe freezing injury. A thermograph was placed inside the freezing chamber and the temperature was recorded. About 5 to 5.5 hours were required to bring the temperatures to the desired level.

Preparation of Samples for Electrical Reading

After freezing, all fifteen plants were removed from the chamber and thawed gradually at room temperature for about five hours. The insulation materials were removed from the cans after thawing. The terminal 6-inch portions of each plant were discarded. From the remaining part three 10-inch lengths of cuttings were taken from each plant. Cuttings were labeled as terminal, median and basal accordingly for each plant. Diameters of the cuttings were measured by calipers graduated in millimeters and recorded as shown in Tables 1-4. Immediately after measuring, the cuttings were placed in polyethylene bags to prevent drying of the twigs.

This procedure was repeated for all three batches of plants treated under the different temperatures as mentioned above and for the control plants. The latter were handled exactly the same as the other trees except that they were not exposed to low temperatures.

After taking the samples from all the plants, the cans were moved to a lath house and placed under conditions favorable for growth. The plants were watered as needed and after about 21 days the survival counts were taken and the results are shown in Table 5.

Direct Reading Method of Measuring Low Temperature Injury

The electrical resistance offered by each twig was measured with a Boyoucous Model C moisture bridge equipped with earphones to detect the null point. Construction and principle of working of this instrument was explained by Campbell (2).

A pair of clamps having parallel acting jaws was used to make contact as shown in Plate I. The lower thick jaw of each had an electrode needle made of steel fixed vertically and permanently which passes through the holes of the upper thin jaw. With the help of a spring the strength between these two jaws was maintained. Electrical wire extensions were fastened internally to these steel needles and the other end was attached to the conductivity bridge. Three such pair of clamps having the distance between the electrodes 0.5, 3 and 6 inches respectively were used to measure the low temperature injury. Each twig thus had three direct readings taken with the needle electrodes at the three distances. These readings expressed in ohms resistance were for 6, 3, and 0.5 inch portions of each treated twig. The end point was noted when the earphone makes the lowest hum. This procedure was repeated for each twig recording the resistance as shown in Tables 1-4.

Exosmosis Method of Measuring Low Temperature Injury

The three twig samples from each of the fifteen plants were measured as described above by the direct reading method. The same twigs were then cut into small pieces each about half an inch long and transferred to beakers. Distilled water about five times the volume of the cut twigs was added. This was repeated for each twig sample from the treated and control plants.

The apparatus used, including the pipette type solution cell, is shown in Plate II.

The beakers containing the water and twigs were kept for about 24 hours in the laboratory. The water solution from each beaker was strained and the residue discarded. The solutions were stirred thoroughly before being drawn into the pipette type solution cell of the resistance bridge for conductance measurement. This solution cell was washed with distilled water each time and also rinsed with a particular electrolyte solution and the electrical conductivity was measured and recorded. The procedure followed for measuring was the same as explained by Dexter (7). Care was taken to prevent air bubbles during filling of the solution cell with electrolytes which otherwise may have interfered with the reading.

The degree of correlation between the two methods as influenced by the size of the twigs was calculated by a regression coefficient analysis as shown in Table 9 and Fig. 4-6.

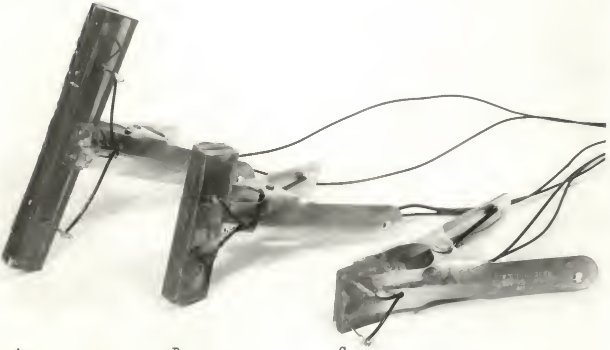
RESULTS AND DISCUSSION

After the sample twigs were taken from each plant the remainder of each plant was shifted to a lath house and placed under conditions favorable for growth. After 21 days, survival counts were taken as shown in Table 5. The

EXPLANATION OF PLATE I

The three sets of clamp type electrode contacts used in determining electrical resistance. Steel needle electrodes are varying distance apart, A 6.0 inch, B 3 inch, and C 0.5 inch.

PLATE I



A B C

EXPLANATION OF PLATE II

Apparatus used in determining electrical conductivity of exosmosed solutions. On the right, Bouyoucos Model C moisture bridge; and on left, pippette type solution cell.

PLATE II


BOUYOUCOS BRIDGE
WOOD & METAL PRODUCTS CO
BLOOMFIELD HILLS, MICH

Table 1. Electrical measurement of twigs of the control peach trees by the direct reading and exosmosis method.

Plant Number	Sample Section Of Twigs	Twig Diameter: In mm.	Electrical Resistance Reading In Ohms — Distance of Electrodes Apart In Inches: 0.5	3	6	Conductivity Reading In Micromohos (Exosmosis Method)
I	Terminal - T	.59	23000	114000	215000	2100
	Median - M	.76	20500	114000	166000	2300
	Basal - B	.93	21000	96000	144000	2600
II	T	.59	24000	126000	220000	2550
	M	.74	16600	102000	192000	2850
	B	.86	22000	194000	215000	1900
III	T	.47	22000	154000	275000	2350
	M	.71	27000	124000	230000	2850
	B	.88	17800	78000	144000	2200
IV	T	.57	22500	164000	225000	2750
	M	.65	26000	126000	225000	2600
	B	.79	23000	94000	200000	2600
V	T	.59	25500	114000	164000	2050
	M	.77	23000	86000	156000	2350
	B	.93	21000	96000	144000	2600
VI	T	.57	21500	138000	150000	2250
	M	.72	28000	104000	196000	2050
	B	.88	17800	78000	144000	2200
VII	T	.46	23500	144000	300000	2350
	M	.58	22000	114000	210000	2600
	B	.72	15400	108000	225000	2750
VIII	T	.52	18000	116000	235000	2450
	M	.65	23000	102000	182000	2200
	B	.82	18600	74000	184000	2500
IX	T	.61	25500	134000	240000	1700
	M	.75	22000	94000	215000	1900
	B	.92	20500	88000	162000	2200
X	T	.66	35000	94000	186000	2600
	M	.79	23000	94000	200000	2600
	B	.89	23500	80000	190000	2450
XI	T	.55	16000	122000	265000	2550
	M	.68	19800	98000	148000	2400
	B	.83	22000	78000	136000	2250
XII	T	.58	21500	112000	164000	2400
	M	.73	21000	84000	188000	2150
	B	.85	18400	74000	138000	2300
XIII	T	.45	19200	144000	325000	2550
	M	.57	18600	122000	230000	3050
	B	.73	21500	124000	210000	2750
XIV	T	.59	17800	112000	164000	2100
	M	.75	18000	96000	184000	2350
	B	.89	23500	80000	190000	2450
XV	T	.45	15400	156000	315000	2650
	M	.57	21500	116000	220000	2900
	B	.69	19800	124000	186000	2950

Table 2. Electrical measurement of twigs of peach trees treated at 8^{o} F. by the direct reading and exosmosis method.

Plant Number	Sample Section Of Twigs	Twig Diameter: In mm.	Electrical Resistance Reading In Ohms — Distance of Electrodes Apart In Inches: 0.5	3	6	Conductivity Reading In Micromohos (Exosmosis Method)
	Terminal - T	.56	7400	21000	47000	720
I	Median - M	.72	9200	24500	47000	1140
	Basal - B	.87	12600	29500	50000	1380
	T	.69	5200	20500	46000	1200
II	M	.76	10800	27500	50000	1240
	B	.85	14600	36500	63000	1620
	T	.51	10200	39000	83000	1240
III	M	.62	7600	31000	67000	980
	B	.73	15600	54000	95000	1800
	T	.42	6000	31500	62000	690
IV	M	.56	7800	25500	57000	780
	B	.69	14200	48000	87000	1560
	T	.52	9800	37000	71000	1200
V	M	.62	12200	32000	65000	1200
	B	.74	13400	41000	72000	1800
	T	.57	8400	20000	49000	840
VI	M	.73	10400	25000	50000	1020
	B	.86	15200	43000	74000	1460
	T	.47	5400	27500	34000	720
VII	M	.69	8200	26500	52000	1120
	B	.79	10000	31500	56000	1480
	T	.73	7400	18200	41000	720
VIII	M	.87	6600	17800	31000	980
	B	1.02	9400	23000	43500	1340
	T	.51	6400	30500	62000	1160
IX	M	.61	9400	32000	67000	1030
	B	.68	16400	61000	114000	1760
	T	.44	5800	28500	46000	800
X	M	.61	9600	24500	48000	880
	B	.77	9800	31500	56000	1420
	T	.66	5800	17800	37000	840
XI	M	.71	7200	27000	61000	980
	B	.82	13600	37000	69000	1440
	T	.61	7600	21000	47000	920
XII	M	.71	8600	22000	50000	980
	B	.81	14600	37000	69000	1460
	T	.57	7800	22000	43000	590
XIII	M	.74	8600	26500	55000	1120
	B	.84	14600	42000	72000	1680
	T	.61	6400	17800	43000	720
XIV	M	.77	9400	21500	27000	880
	B	.85	16400	41000	73000	1460
	T	.61	6400	24000	50500	880
XV	M	.73	10600	31000	52000	1380
	B	.83	14000	38000	78000	1680

Table 3. Electrical measurement of twigs of peach trees treated at -10° F. by the direct reading and exosmosis method.

Plant Number	Sample Section Of Twigs	Twig Diameter In mm.	Electrical Resistance Reading In Ohms — Distance of Electrodes Apart In Inches 0.5	3	6	Conductivity Reading In Micromohos (Exosmosis Method)
	Terminal - T	.51	5300	24500	58000	840
I	Median - M	.61	5600	27500	54000	1180
	Basal - B	.69	7700	29000	60000	1300
	T	.64	5200	24000	42000	820
II	M	.78	5200	20500	41000	680
	B	.98	7200	22000	46500	1260
	T	.48	9800	33000	81000	1140
III	M	.62	6400	35000	73000	1020
	B	.71	12800	49000	95000	1640
	T	.72	6800	22500	54000	880
IV	M	.82	7300	20000	41000	800
	B	.96	7200	25000	45500	1260
	T	.43	6800	42000	79000	1080
V	M	.57	5300	36000	72000	1200
	B	.65	13400	45000	97000	1340
	T	.65	5700	22500	40500	840
VI	M	.81	6600	20500	40500	1020
	B	.96	7200	23000	48500	1240
	T	.56	5000	22500	55000	660
VII	M	.86	7600	20500	51000	860
	B	1.01	8300	21000	44500	1320
	T	.53	6400	24000	54000	980
VIII	M	.71	7800	25000	55000	780
	B	.83	7900	23500	52000	1170
	T	.51	5400	30000	66000	1160
IX	M	.62	8600	28000	53000	900
	B	.74	8100	30500	53000	1460
	T	.54	7200	38000	82000	990
X	M	.66	10000	30500	61500	1140
	B	.78	9800	31500	57000	1220
	T	.61	5800	20500	45000	810
XI	M	.76	6600	25500	46500	1170
	B	.84	9800	24000	48500	1380
	T	.58	14600	38000	84000	970
XII	M	.79	10400	31500	50000	1140
	B	.89	7400	25000	45500	1160
	T	.52	5700	36000	73000	1040
XIII	M	.62	7600	31000	56000	870
	B	.74	9800	34000	62000	1420
	T	.45	4600	31000	63000	840
XIV	M	.58	7800	38000	71000	800
	B	.75	9200	36000	67000	1360
	T	.47	6200	32500	71000	1120
XV	M	.57	10200	34000	60500	1120
	B	.68	12600	37000	67000	1620

Table 4. Electrical measurement of twigs of peach trees treated at -20° F. by the direct reading and exosmosis method.

Plant Number	Sample Section Of Twigs	Twig Diameter: In mm.	Electrical Resistance Reading In Ohms — Distance of Electrodes Apart In Inches: 0.5	3	6	Conductivity Reading In Micromohos (Exosmosis Method)
I	Terminal - T	.47	4400	29000	39000	1240
	Median - M	.65	4000	27000	46000	1300
	Basal - B	.79	6400	23000	39500	1200
II	T	.57	4000	24500	48000	1020
	M	.68	5300	24500	51500	1120
	B	.82	6400	32000	40000	1220
III	T	.55	3200	26000	59000	1060
	M	.73	6200	26000	44500	1220
	B	.84	6200	25500	54000	960
IV	T	.48	4000	27500	69000	1040
	M	.64	3700	34000	54000	1180
	B	.71	6400	30000	50500	1480
V	T	.47	6600	46500	94000	1440
	M	.69	6200	41500	83000	1540
	B	.73	12200	51500	94000	1740
VI	T	.74	3200	21000	39500	940
	M	.87	3100	19400	32000	980
	B	1.02	4800	18000	26500	1000
VII	T	.43	3900	23000	20500	920
	M	.61	7600	24000	48000	1040
	B	.74	6800	28500	43000	1400
VIII	T	.63	4400	21500	31000	1140
	M	.77	4100	21500	28000	1100
	B	.96	5100	22500	31000	1270
IX	T	.64	3600	28000	54000	1140
	M	.69	8400	27000	50000	1340
	B	.82	7600	34000	53000	1470
X	T	.41	5000	33000	66000	980
	M	.56	8000	30500	56000	1300
	B	.72	9800	33500	62000	1780
XI	T	.69	3800	22000	36000	1180
	M	.85	5100	20000	28500	1080
	B	1.04	4800	18600	28500	1200
XII	T	.64	5100	26000	43000	1240
	M	.76	5900	25500	32000	1420
	B	.91	6400	22000	33000	1240
XIII	T	.66	5200	24500	47000	1260
	M	.77	5400	20000	35000	1180
	B	.89	6200	24000	31500	1240
XIV	T	.52	5600	26000	59000	1280
	M	.62	3800	29000	47000	1350
	B	.74	6800	28500	43000	1400
XV	T	.49	2700	20500	45000	700
	M	.63	5800	21000	40000	1000
	B	.77	5600	27500	40500	1450

data from the direct reading (resistivity) method were subjected to an analysis of variance shown in Tables 6-7. The correlation coefficient of electrical readings for both methods used in this investigation are presented in Table 9.

Three twig samples from each of the fifteen trees subjected to different temperature treatments were believed to be sufficient (Table 6). Electrical resistance as measured with the Model C Bouyoucos Bridge in the direct reading method is given in Tables 1-4.

The data reveals that the control plants offered greater electrical resistance than the trees subjected to different freezing temperatures, Tables 7-8. In the exosmosis method the average electrical conductivity reading was converted into electrical resistance reading by the formula $R = \frac{C}{K}$ where K denotes the constant of the pipette type solution cell shown in Plate II. Numerically this value was two. When comparing twigs with different diameters, there was no proportionate increase or decrease in electrical readings found either with an increase or decrease in diameter with either the direct reading or exosmosis method (Tables 1-4 and 9). It can be seen in Tables 1-4 that with an increase in distance between the electrodes increased electrical resistance was found with the direct reading method.

Table 5. Percentage survival count taken 21 days after treatment for both treated and control plants.

Temperature Treatment	Number of Plants Treated	Number of Plants That Survived Treatment	Percentage Survival
Control	15	15	100.0
8° F.	15	14	93.3
-10° F.	15	15	100.0
-20° F.	15	14	93.3

Table 5 shows the survival of trees 21 days after low temperature treatment. The control plants and those receiving the -10° F. treatment showed 100 per cent survival. The degree of damage of tissues may depend upon the amount of low temperature under which trees are treated as well as on other factors not measured in this investigation. Each tree in the lots of fifteen from 8° F. and -20° F. treatments found to be dead, may have died due to prior treatment. Possibly the single tree in each treatment which died may have been damaged due to excessive root pruning at the time of planting in the can or to drying out. Under conditions of this experiment, the majority of the Golden Jubilee peach trees survived the low temperature of -20° F.

Table 6. Analysis of variance of electrical resistance (in ohms) of peach trees subjected to four different temperatures measured by the direct reading method at three different electrode distances.

Source	D.F	Sum of Squares	Variance
Temperature	3	644878852826	214959617608**
Plants:Temperature	56	41967363822	749417211**
Samples:Plants	120	35634346667	296952889
Distances	2	578371241148	289185620574**
Distances X Temperature	6	288615072841	48102512140**
Distance X Plant:Temperature	112	23100509345	206254548
Distance X Sample:Plant	240	1424890143351	5937042263

**Highly Significant

Table 6 shows the different sources of variance involved in low temperature injury measurement with the direct reading method.

Temperature variance was found highly significant when tested at the one per cent level in the direct reading method. With each decrease in temperature, there was a consistent reduction in the electrical resistance reading. Evidently the different temperature treatments caused proportionate changes in the electrical resistance. There were significant differences in electrical resistance between groups of trees receiving different temperature treatments. As seen in Tables 2-4, in general the lower the temperature treatment, the lower the electrical resistance offered by plants.

All plants treated at the same temperature showed highly significant variability in electrical resistance readings at the one per cent level. However, the variability among temperatures was greater than that among plants of the same temperature. This suggests that temperature had a greater effect on the variability of electrical resistance measurement than the other factors affecting plant behavior for such measurements. A highly significant variability for the distance between the electrodes was noticed. This indicates that a proportionate increase in the electrical resistance was found with an increase in electrode distance. This suggests the type of damage and the electrical property of the plant cell to be uniform throughout the twig length.

The interaction between a given electrode distance and each different temperature was found highly significant at the one per cent level. The electrical resistance when measured at different electrode distances varied with different temperature treatment. This shows certain changes in the electrical resistance of tissues when exposed to experimental temperature treatments.

The interaction between electrode distance and plants treated with the same temperature was found to be non-significant. This suggests that the different temperature treatments caused similar effect on resistivity measurements upon every plant at a given electrode distance. This can be true if the type of damage is uniform throughout the twig length.

Table 7. Average electrical resistance (in ohms 10^2) of peach twigs measured at 0.5, 3 and 6 inch electrode distances on control plants and plants treated at 8° F., -10° F. and -20° F.

Distance Between Electrodes	Control Trees	8° F.	-10° F.	-20° F.	Average of Distance
0.5 inch	214.74	99.24	78.20	55.29	111.86
3.0 inch	1085.78	303.24	293.33	268.67	487.76
6.0 inch	1999.33	583.44	591.67	465.67	909.28
Average of Temperature	1099.94	327.64	321.07	263.21	

L.S.D. of temperature at 5% level 168.49
L.S.D. of electrode distance at 5% level 117.80
L.S.D. of electrode distance X temperature at 5% level 765.49

Table 7 shows the average electrical resistance of peach twigs from each batch of fifteen trees treated at 8° F., -10° F. and -20° F. together with twigs from control trees measured at 0.5, 3 and 6 inch electrode distances. The L.S.D.s for electrode distance, temperature and the interaction between them at the five per cent level are also given. The differences in average resistance in ohms between trees of various lots subjected to the low temperature treatments were not significant. However, the average resistance readings of the twigs from trees subjected to the low temperatures were all significantly lower than for the control twigs. Since the resistance readings of the treated trees failed to differ significantly, the critical temperature responsible for the changes in electrical reading in the tissues was not determined. These changes in electrical property of the tissues occurred somewhere between 8° F. and 38° F. As seen in Table 7, the electrical readings varied significantly between each two electrode distances regardless of temperature treatment. The electrical resistance was found proportionately higher with increase in electrode distance used.

There were significant interactions between electrode distance and plants subjected to different temperatures in control trees, but not in any of the treated plants. The electrical resistance in twigs of control trees measured at 0.5, 3, and 6 inch electrode distances was found to be consistently higher than for twigs from the trees treated at 8° F., -10° F. and -20° F. These lower readings can be assumed to reflect relative injury to the twigs from the low temperatures. Therefore one can assume that the critical temperature which caused a drop in electrical resistance of the twig tissues lay between 8° F. and the temperature used for control trees (38 ± 3° F.). By comparing electrical resistance readings with the percentage survival count of all the experimental plants, it can be concluded that most of the treated plants were damaged but not killed.

Table 8. Average electrical resistance (in ohms) determined by the exosmosis method of the twigs from control peach trees and from trees treated at 8° F., -10° F., and -20° F.

Plants With and Without Treatment	Control Trees	8° F.	-10° F.	-20° F.
Average Electrical Resistance	1213.89	581.00	545.55	608.66

Table 8 shows the average electrical resistance of the electrical readings as determined by the exosmosis method of the fifteen control trees and each batch of fifteen trees treated at 8° F., -10° F. and -20° F. temperatures. The electrical resistance of the exosmosed solution of twigs from the control tree was found to be much higher than for the treated ones. There was no obvious proportionate increase or decrease in electrical resistance when the temperature was lowered. This suggests the exosmosis method of measuring low temperature injury to be highly complicated. Trees treated at -20° F.

temperature offered more electrical resistance than the trees treated at 8° F. and -10° F., but more electrical resistance was noted in twigs from trees treated at 8° F. than from those treated at -10° F. Because of the complicated factors involved in the exosmosis method of injury measurement the reasons for these results are still unknown.

Table 9. Correlation coefficients of the direct reading method used for measuring low temperature injury at 0.5, 3 and 6 inch electrode distances with exosmosis method and twig diameter.

	Direct Reading At 0.5" Distance	Direct Reading At 3" Distance	Direct Reading At 6" Distance	Exosmosis	Diameter
Direct Reading At 0.5" Distance		.866473	.847263	.851635	.043011
Direct Reading At 3" Distance			.966680	.896385	-.188464
Direct Reading At 6" Distance				.873430	-.211326
Exosmosis					.070965
Diameter					

In an effort to evaluate and compare the two methods of measurement, correlation coefficients were calculated as presented in Table 9. The correlation coefficient of electrical readings by both methods with twig diameter are also shown.

A significant positive correlation between all electrode distances when the direct reading method was employed can be seen from this table and also in Fig. 1-3. The significant positive correlation for electrode distances emphasizes that any one of the three electrode distances would have produced essentially the same results, provided all the factors for measuring

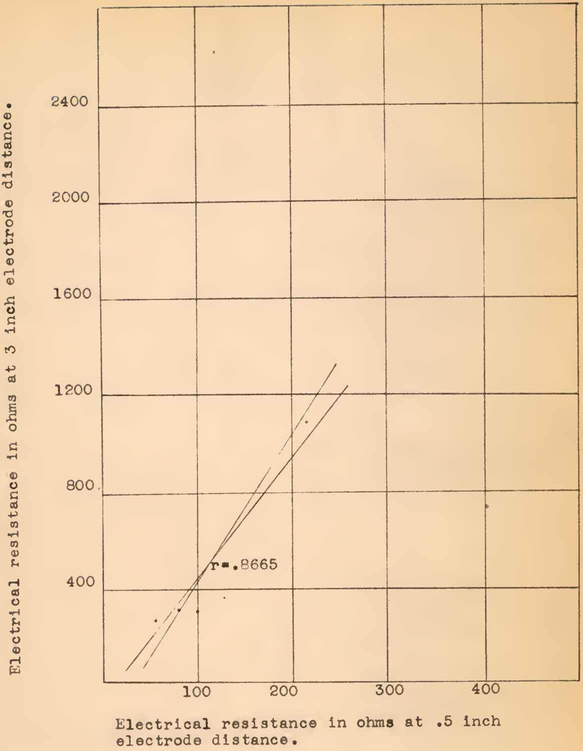


Fig. 1. Correlation coefficient of electrical resistance from each peach twig samples measured at .5 inch and 3 inch electrode distances.

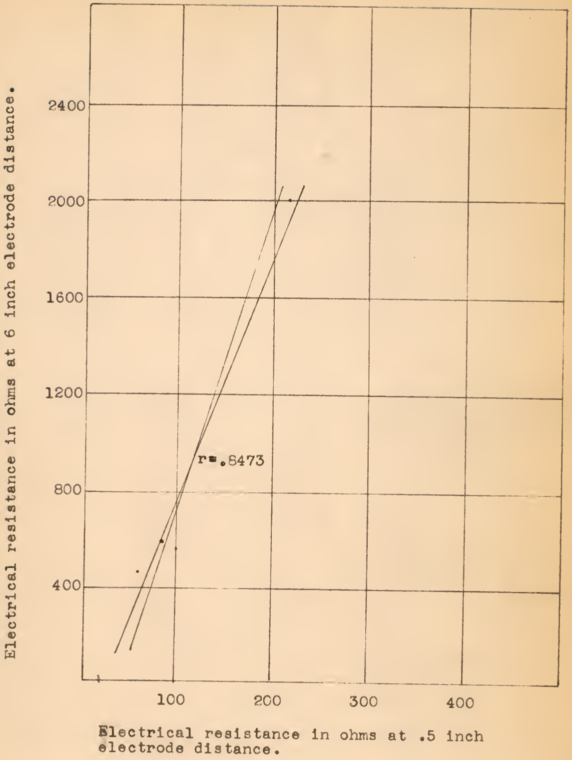


Fig. 2. Correlation coefficient of electrical resistance from each 60 peach twig samples measured at .5 inch and 6 inch electrode distances.

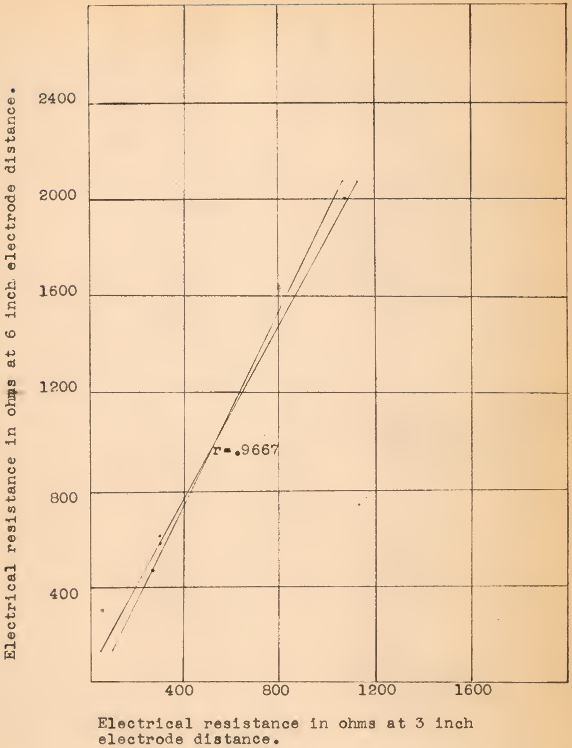


Fig. 3. Correlation coefficient of electrical resistance from each 60 peach twig samples measured at 3 inch and 6 inch electrode distances.

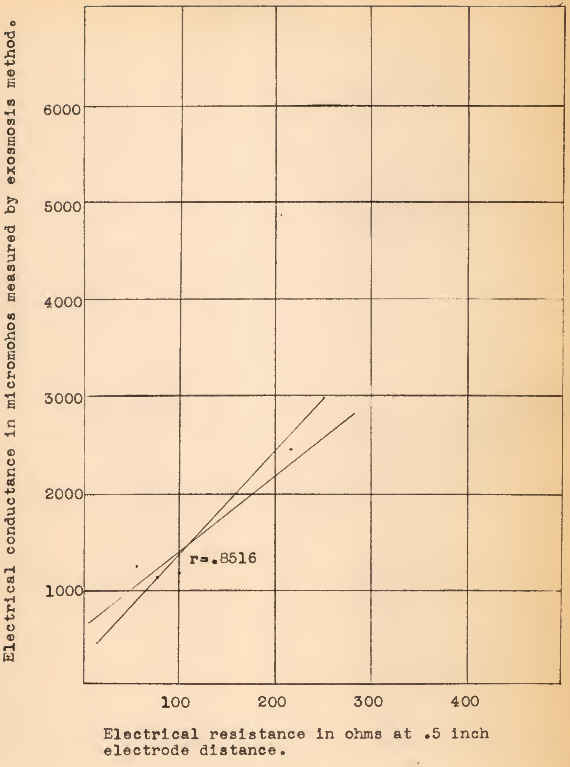


Fig. 4. Correlation coefficient of resistance and conductance from the same 60 peach twig samples measured at .5 inch electrode distance and exosmosis method.

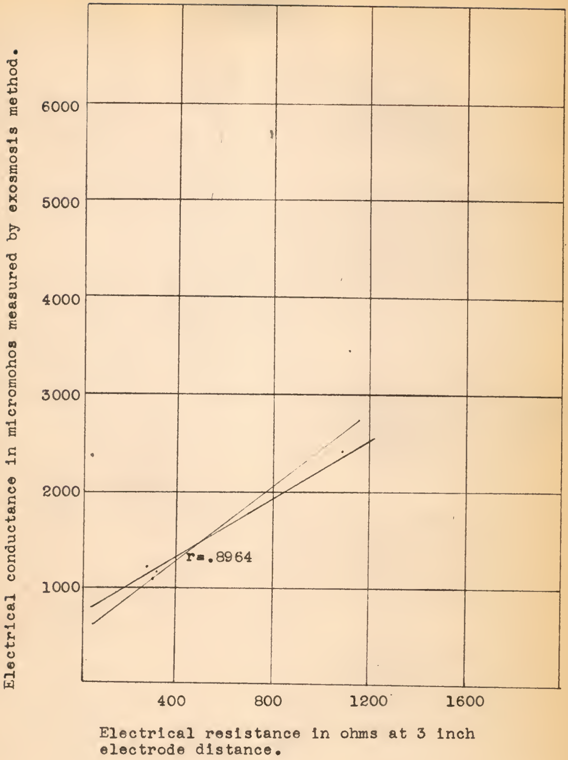


Fig. 5. Correlation coefficient of resistance and conductance from the same 60 peach twig samples measured at 3 inch electrode distance and by exosmosis method.

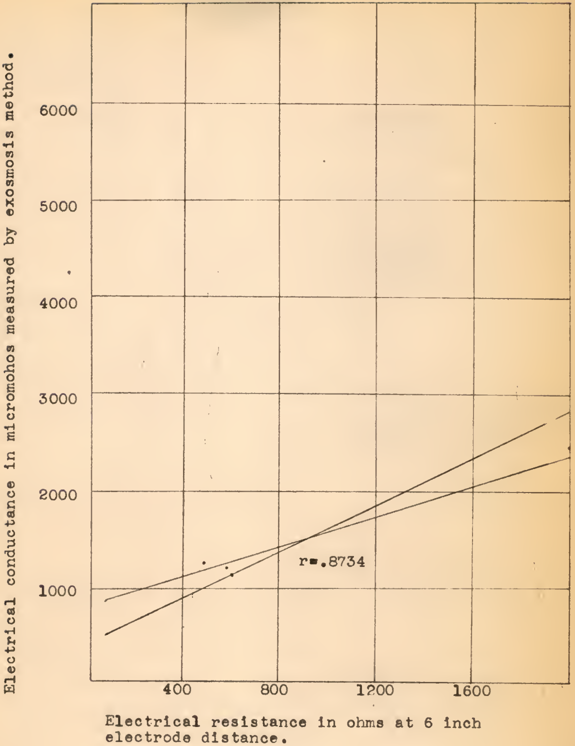


Fig. 6. Correlation coefficient of resistance and conductance from the same 60 peach twig samples measured at 6 inch electrode distance and by exosmosis method.

electrical resistance were the same. A partial difficulty in the procedure of measuring the electrical resistance at 0.5 inch distance was noticed. Since the electrodes were very close together, the twigs split apart tending to increase the resistance reading when compared to twigs not split apart. Also the splitting would be a source of injury to intact twigs on the trees and therefore would be undesirable. When the electrode distance was increased the consistency in electrical resistance also increased. At the same time with an increase in distance between electrodes, difficulty in handling the apparatus was observed. Therefore medium electrode distances of 3 inches may be preferable over 0.5 or 6 inches to measure the electrical resistance of woody plants.

Significant positive correlation was observed between the exosmosis method and direct reading method regardless of electrode distance (Fig. 4-6). Hence these two methods were found to be very accurate and reliable in the electrical measurements of low temperature exposed woody trees. At the 3 inch electrode distance the direct reading and exosmosis methods had a value of $r = .89$ correlation which is a higher degree of relationship in electrolytic measurement than the correlation between 0.5 inch and exosmosis or 6 inch and exosmosis, suggesting a closer relationship between electrical measurement of the direct reading method at 3 inch electrode distance and the exosmosis method.

There was no consistency in the results or significant relationships observed between twig diameter and electrical readings. A negative non-significant correlation of twig diameters at the 3 inch and 6 inch electrode distance was noted. Also the twig diameters showed no consistent relationship to electrical conductance as measured by the exosmosis method. This suggests all the trees that survived to be well matured and in dormant condition.

The major advantage in the direct reading method was that it could be used to measure the injury without seriously damaging the tissues as explained by Filinger (11). By clamping the pair of steel electrode needles directly into the twigs, the changes in electrical resistance of one year old peach trees were measured, hence the name "direct reading method". The procedure for testing winter hardiness by this method is simple and rapid; therefore field hardiness investigation can be carried on without damaging the tissues. In addition, twig survival probability after winter exposure also can be predicted. The exosmosis method has the disadvantages of being tedious and time consuming. Further, twig samples that are tested must be removed from the trees and are destroyed in the process.

SUMMARY

1. The direct reading and exosmosis methods were found to be significantly correlated suggesting both methods to be equally accurate and reliable in testing for low temperature injury.

2. The electrical resistance offered by twigs of the control peach trees was much greater than for those receiving low temperature treatments in both the direct reading and exosmosis methods.

3. There was no significant correlation between twig diameter and electrical readings as measured by either the resistivity or conductivity methods.

4. With each decrease in temperature, there was a consistent reduction in electrolytic reading as determined by both methods.

5. The three electrode distances compared in the direct reading method used in this investigation were found to be equally accurate in measuring low

temperature injury.

6. In the direct reading method, as the distance between the electrodes increased, the electrical resistance offered by peach twigs also increased.

7. The analysis of variance indicated greater variability in electrolytic resistance due to temperature than from differences in the plants. The drop in electrical resistance was thought to reflect low temperature injury. Interaction between electrode distances and plants under temperature treatment was not significant.

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A COMPARISON OF METHODS FOR THE DETERMINATION OF LOW TEMPERATURE INJURY TO PEACH TREES

by

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The purpose of this study was to determine whether there was any significant correlation between the direct reading method and the exosmosis method of measuring the relative effect of low temperature on Golden Jubilee peach trees. The correlation of low temperature effects with twig diameter was also studied. In the direct reading method an effort was made to determine if the distance between the electrodes affected the electrical resistance measurements.

In the direct reading method, the procedure of measuring the electrical resistance from the twigs of trees subjected to different temperatures was similar to that reported by Filinger and Cardwell (1941). The method used to measure the electrical conductance of an exosmosed water solution in which portions of twigs were placed was essentially the same as reported by Swingle (1932).

One-year old dormant, mostly single whip peach trees were used. Extensive roots were pruned and the trees planted in gallon cans containing a soil mixture of 3:1 sandy loam and peat moss respectively. The plants were watered sufficiently and hardened for about 30 days at the temperature of $38 \pm 3^{\circ}$ F. with an average light intensity of 32.8 watts per square foot for about ten hours each day.

Batches of fifteen plants were treated at three temperatures 8° F., -10° F., and -20° F. separately after the cans were wrapped with an insulating material to protect the root system. After 5 to 5.5 hours they were removed from the freezing chamber and thawed gradually at room temperature for about 5 hours. After discarding the terminal 6 inch portion, three twig samples of about 10 inches were taken from each tree and their average diameter was recorded. The cuttings were labeled and placed in polyethylene bags to prevent drying out. The same procedure was followed to prepare the twigs from the control trees except that the plants were not exposed to low temperatures.

After the twig samples were removed, the cans with the remainder of each plant were placed under conditions favorable for growth and after 21 days the percentage survival count was taken.

Three different clamps having the needle electrodes 0.5, 3 and 6 inches apart were clamped one at a time directly into each twig and the electrical resistance offered by each was measured with the Bouyoucos bridge.

The same twigs were cut into one-half inch pieces and placed in a beaker to which distilled water about five times the volume of the cut twigs was added. The beaker with the contents was kept for about 24 hours at room temperature after which a portion of the water solution was drawn into the pipette type solution cell and the electrical conductance was measured.

From the data of both the resistivity and conductivity methods it was found that twigs from the control tree offered more electrical resistance than the twigs of trees treated at different low temperatures.

The data were analyzed statistically. With a decrease in temperature a proportionate decrease in electrical resistance was found with the direct reading method. The critical temperature which influenced the changes in electrical resistance was assumed to be somewhere between the temperature of control trees 38° F. and the treatment 8° F. With each increase in distance between the electrodes, a proportionate increase in the resistance reading was observed. However, a high positive correlation was found to exist between all the direct readings regardless of distance between electrodes.

Also a significant positive correlation was observed between the exosmosis method and direct reading method regardless of electrode distance. There was no significant correlation between twig diameter and electrical readings as measured by either the resistivity or the conductivity methods. Both methods used to measure the electrical reading of twigs of trees exposed to low temperature were found to be accurate and reliable.