ANATOMICAL ASPECTS OF SOME ORNAMENTAL PLANTS SUBJECTED TO SPECIFIC TRANSPLANTING TECHNIQUES

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INTRODUCTION

Nursery grown trees and shrubs usually make little growth the first year after transplanting. Numerous observations have shown that this lack of growth may be caused by the difficulty of re-establishing the root system (Emerson and Hildreth, 1933; Pridham, 1941; Swartley, 1941; and Gossard, 1942).

Various hormone preparations have been shown to increase root quantity on plants (Harrison, 1937; Mitchell, 1938; Smith, 1938; Gossard, 1942; and Doran, 1952). On some plants roots have been produced more abundantly, as well as more rapidly, when the hormone substances were applied to the vegetative parts. Swartley (1941) and Nagato and Sato (1952) have reported more rapid root development on already rooted plants which were soaked at transplanting time with hormones.

A number of plants are marketed with a heavy bulk of soil surrounding their roots. Since soil weighs about 100 pounds per cubic foot, the plant is difficult to handle, and the root system is frequently broken and bruised, causing difficulty in its rehabilitation into the landscape. Miller, Neilson and Bandemer (1937), and Gartner, O'Rourke and Hamner (1949) have successfully transplanted plants without having their roots surrounded by soil. This method of transplanting has not been a general practice for larger vascular plants, but is often used for young nursery stock (Thompson, 1940).

Following transplanting, it is believed that root and top desiccation is one of the major causes of death.

The present investigation was undertaken to study (1) the anatomical aspects of vacuum treatment as a possible method to reduce the weight of transplanting stock; (2) effects of indoleacetic acid forced into the plant; (3) the effects of the use of polyethylene film as a covering to prevent desiccation of *Thuja occidentalis* cv. nigra during the late winter and early spring.

LITERATURE REVIEW

Transplanting:

Even though transplanting of ornamental plants has been a commonplace operation for years, reports of high mortality rates of transplanted stock are common. Gregory, Imle, and Camacho (1953) reported high losses despite the fact that great care was given to digging and planting of *Hevea brasiliensis*. In addition, they defoliated and pruned the crown to balance the reduced root system.

Transplanting experiments involving various species of evergreens by Emerson and Hildreth (1933) resulted in mortality rates as high as 100 per cent. They attributed the loss to (1) climatic factors favorable to high transpiration; (2) the necessity of moving the plants in full foliage; and (3) the slow rate of root regeneration.

Transplanting seriously reduced growth of *Quercus palustris* (Pridham, 1941). It was concluded that the reduced growth could not be attributed entirely to root and top pruning, but that the season, soil, and the care in handling were important factors.

According to Whitten (1919), the majority of investigators have recommended that plants be moved in the spring. Thompson (1940) stated that some authorities recommended fall, some winter, and some spring, and still others pointed out that plants may be moved successfully at any season of the year. He stated, further, that although plants were moved every month of the year, greater injury would occur if the plant was disturbed during its period of rapid growth.

In an effort to reduce injury and to lengthen the transplanting season, Miller, Neilson and Bandemer (1937) reported that by spraying the tops with a wax emulsion they were able to transplant the following bare-root specimens successfully in July: Picea Abies, Acer palmatum cv. dissectum, Ulmus americana, Populus nigra cv. italica and Syringa vulgaris.

Transplanting experiments conducted in 1949 by Gartner, O'Rourke and Hamner made use of a protective covering of vinyl resin latex sprayed on the foliage. Both sprayed and unsprayed trees of Picea alba transplanted in June declined in general vigor, but the treated trees were always in better condition than a comparable number of controls. They found that all deciduous plants did not respond alike. Bechtel's flowering crab (Malus ioensis) and lilac (Syringa Spp) showed symptoms of growth inhibition, accompanied by the abscission of some leaves. It was indicated that cessation of growth as a result of transplanting was prolonged by the application of the plastic resin, but that the plant usually regained any loss of growth by a more vigorous production of new shoots after growth was resumed.

Another method of overcoming the loss of vigor and to produce rapid reestablishment of the plant was the use of starter solutions. Stair and Hartman (1939) found that by using nutrient solutions in the transplanting water for tomato (Lycopersicum esculentum) plants, they were able to increase slightly the early yields. They were unable to show significant differences between treatments for total yields. Similar results were obtained by Sayre (1939).

It was believed by Thompson (1940) that in general a well-balanced fibrous root system was probably the most important single factor for success in transplanting. One method of increasing the transplanting bulk of roots of a tree was root pruning well ahead of the time of transplanting. To demonstrate the feasibility of this procedure, Faulkner (1953) root pruned one-year-old "Corsican" pine seedlings. He found that root pruning markedly increased the production of the fibrous roots in the soil above the point of root severance.

Neff and O'Rourke (1949) presented data which indicated that transplanted tung trees (Aleurites Fordii) tended to make top growth, followed by root growth. They concluded that proper cultivation exerted perhaps the greatest influence in minimizing the decline in the growth rate as deficient moisture markedly checked top growth.

According to Goff (1897), failure of a tree to resume growth following transplanting was not always caused by insufficient moisture in the soil. Water pressure within the bud, supplied largely through root pressure caused the expansion of the leaves. The undisturbed plant maintained the root pressure by means of living root hairs. By attaching one end of a rubber hose to a root 1/4 to 3/8 inches in diameter, the opposite end of which was connected to a flask of distilled water above the height of the tree, water under pressure was supplied to the tree. The buds of the treated trees expanded more rapidly than those of the untreated trees. Following soil irrigation, all trees began growth.

Growth Hormones:

Gustafson (1942) has referred to his earlier publication of 1936, in which he disclosed that indoleacetic, indolepropionic, indolebutyric, and phenylacetic acids induced seedlessness of mature fruits by placing lanolin paste mixture on the style of unpollinated flowers of *Lycopersicum esculentum*, *Petunia hybrida*, and *Salpiglossus variabilis*. He stated further that since that time many crops have been added to the list of parthenocarpic fruit production as a result of treating with synthetic growth substances.

Hormones have since been used to achieve other results. Zimmerman and Hitchcock (1951) has listed (1) the vegetative propagation of plants by treatment of the cuttings and scions; (2) the prevention of pre-harvest drop of fruit, particularly apples and oranges; (3) the increase of fruit set and induction of parthenocarpic fruits; (4) the inhibition of buds to prevent potato and onion sprouting; (5) the inhibition of fruit tree buds or prolonging dormancy to prevent loss by late frosts; (6) the inhibition of buds of nursery stock to prevent top growth until the young plants are established; (7) regulating the flowering of various crops; (8) the premature defoliation of plants; (9) the prevention of abscission as contrasted to defoliation; (10) the chemical thinning of fruit; and (11) the use of chemicals as selective weed killers. He foresaw the future need of bud inducing chemicals in order to facilitate propagation of plant parts where they were lost or where buds had not appeared.

One of the more popular methods of application of hormone substances has been to mix it with lanolin (Howlett, 1939; Gustafson, 1940; Asen and Hamner, 1953). The vapor method of application was introduced by Zimmerman and Hitchcock (1944). A similar method is as a spray emulsion used by Withrow (1945). Cholodny (1936), Stier and duBuy (1938), and Thimann and Lane (1938) applied hormones by soaking the seed in an aqueous solution containing the growth regulator. Harrison (1937) and Block (1938) dipped the plants to be treated in a solution containing the hormones. The talc method, which is described by Doran (1952) is used extensively by plant propagators. Applications of growth regulators were made to the soil by Randhawa and Thompson (1948) and Swartley (1941). Before transplanting rice seedlings (*Oryza sativa*) Nagato and Sato (1952) soaked them in a solution containing naphthalene acetic acid.

Localized application of hormone substances was used by Romberg and Smith (1938) in transplanting pecan (*Carya illinoensis*). This was accomplished by first soaking toothpicks in a solution containing the growth regulator. Holes were bored into the roots and a toothpick plug inserted.

Watson (1948) and Mitchell and Brown (1946) injected measured amounts of 2, 4-dichlorophenoxyacetic acid into leaf tissue from a hypodermic needle.

Conflicting reports have been recognized of the response of plants to applications of hormones. Higher concentrations of growth regulators have caused a general inhibition of growth, a modification of parts as well as loss of the smaller flower buds (Zimmerman and Hitchcock, 1944). In an earlier investigation they found that for a few days following treatment the buds of *Hibiscus syriacus* grew at equal rates in a measured period. The uppermost shoots soon after application grew more rapidly, inhibiting the lower buds. Removing the distal tip of the stem, stimulated growth of the inhibited shoots. Negative results were obtained by using a concentration of 0.5 per cent indoleacetic acid to induce root formation on intact plants of Tradescantia valida and Zebrina pendula (Dorfmuller, 1938). A slight reduction in fresh weight of Tagetes Spp., and a slight increase in weight of Phaseolus vulgaris var. Black Valentine was reported by Mitchell (1938). The treatment of one ppm indoleacetic acid had no noticeable influence on the time of flowering.

Zimmerman and Hitchcock (1944) likewise were able to demonstrate that even though hormone-treated Lycopersicum esculentum fruit grew faster than the control, there was a difference among cultivars. The cultivar Globe was more sensitive than the cultivars Marglobe or Bonny Best. The development of photosynthetic area and time of flower of cultivar Comet has been hastened by soaking the seed in a hormone solution under greenhouse conditions (Thimann and Lane, 1938). Seedlings of Oryza sativa soaked in a solution of naphthalene acetic acid before transplanting were shown to have their root growth retarded for one or two days (Nagato and Sato, 1952). Following this period root growth was hastened and this effect was diminished gradually under favorable growing conditions which caused faster production of tillers, heading, and heavier fruiting, than if the plants had not been treated. Root production on pecan, although not named as such in the publication, it was assumed to be Carya illinoensis, was substantially increased by localized applications of indolebutyric acid by inserting well-soaked toothpicks into the roots (Romberg and Smith, 1938 and Gossard, 1943). Rooting response was greater in the younger trees, with the higher concentrations producing the greatest response (Romberg and Smith, 1938). Harrison (1937) obtained similar results when treating Iresine lindenii with indoleacetic acid. Root and stem development has been shown to have been stimulated by hormone treatment on young plants of some species of the following genera: Lonicera, Malus, Juniperus, Coreopsis, Viola, Incarvillea, Lycopersicum, and Callistephus (Swartley, 1941).

Zimmerman and Hitchcock in 1942, using triiodobenzoic acid caused the loss of the apical dominance of Lycopersicum esculentum and produced an abnormally large number of axillary leafy branches. Using the same material, Asen and Hamner (1953) have increased the number of basal shoots on Rosa hybrida cv. Better Times.

Thimann (1937) reported that very low concentrations of auxins have accelerating effects on the vegetative portions of the plants. Phaseolus vulgaris cv. Calapproved increased in dry weight, absorption of water and directional shifts of large amounts of nitrogen and carbohydrates from leaves and cotyledons in comparison to the controls (Stuart, 1938). Inhibition by the presence of the growing apex of Phaseolus vulgaris was demonstrated by Snow, 1925. When the apex was removed or prevented from growing, one or more proximally located axillary buds began growth. Vitis sp. cuttings of 31 varieties, after treatment with various growth regulators, showed variable results of rooting (Harmon, 1943). Inconsistent and non-conclusive results were reported by Leibundgut (1950) on growth of roots and shoots of hormone-treated forest tree seeds.

The tap root and lateral roots of Hevea brasiliensis were treated with naphthaleneacetic acid in concentrations of 0.5 per cent and 1 per cent by Gregory, Imle, and Camacho (1953). From this application they concluded that NAA helped increase the degree of survival of the transplanted trees.

A variety of responses of the yield of fruit may be expected from hormone treatment. Stier and duBuy (1938) increased the early and total yields of marketable *Lycopersicum esculentum* fruit by soaking the seed in various hormone solutions. Seed of *Avena sativa*, which was soaked in a solution of indoleacetic acid before sowing, produced plants on which the yield of seed per plant was increased by 55 per cent (Cholodny, 1936). A concentration of 0.25 mg of 4-phthalimido-2, 6-dimethyl pyrimidine per litre was found to increase the early yields of *Lycopersicum esculentum* when compared with treatments which are generally accepted methods of hardening (Learner, 1952). There was also an increase in the total number of ripe fruit harvested.

Withrow (1945) treating *Lycopersicum esculentum* cultivars Long Calyx and Michigan State Forcing with growth regulators and growing them in the greenhouse from April to July, reported the formation of more fruit on the early clusters, but found little difference in total production.

Field-grown plants produced practically no difference in yield after having been treated (Zimmerman and Hitchcock, 1944). The same plant watered at transplanting time with a concentration of 50 mg naphthaleneacetic acid in 50 gallons of water suffered a reduction of early fruit (Sayre, 1938). When five times as much hormone was applied, there was an even greater delay on date of maturity and a lower yield. Sayre concluded that hormones were more effective in very dilute concentration. It would seem that the chief benefit to be derived from the use of hormones on tomatoes would be the production of parthenocarpic fruits.

Morphological and histological differences occurred in plants which had been treated with hormones. Responses differed with the species of the plant, and the concentration of the hormone. A general review of the literature to the present has been made by Beal in 1951. Hormones applied to roots, stems, leaves, flowers or fruits were readily absorbed by the plants (Mitchell, 1951). It had been pointed out previously by Mitchell and Martin, 1938, that in *Phaseolus vulgaris* marked morphological differences occurred following treatment with indoleacetic acid. Indoleacetic acid, when applied to *Lycopersicum esculentum* did not inhibit the development of buds below the treated area. Responses were similar to those of *Phaseolus* sp., except in *Lycopersicum esculentum*, which was explained on the basis of the presence of internal phloem in this plant (Borthwick, Hamner, and Parker, 1937).

In response to various growth regulators *Hibiscus syriacus*, according to Zimmerman and Hitchcock (1940) usually produced new adventitious buds associated with the xylem points which extended into the bark. *Populus generosa* did not respond in a similar manner.

The growth of axillary buds of *Tradescantia fluminensis* was suppressed by dipping the plants in an aqueous solution of indolebutyric acid (Block, 1938). He also reported that in the rapidly elongating parts of the shoot, short thick segments were formed in the basal part of the internode. The differentiation processes were initiated prematurely and more vigorously. This resulted in the formation of numerous, fasciated, internal roots as well as nodal vascular elements.

Cooper (1938) concluded that indoleacetic acid controlled the movement of substances in the cuttings. Removal of the base of a hormone treated cutting, followed by a second treatment, seemed to destroy the effect of the treatment.

Desiccation:

Meyer and Anderson (1952) have cautioned the reader not to confuse winter killing, resulting from desiccation with cold injury, resulting from low temperatures, which is a different phenomenon.

At temperatures slightly above freezing, the rate and amount of water movement in plants was greatly reduced (Kramer, 1940).

Thompson (1940) attributed injury to certain deciduous and evergreen plants to desiccation by winds in late winter and early spring. He explained that the winds caused excessive transpiration from the foliage and twigs of a number of different plants before the roots were able to absorb sufficient water to replace that lost from the leaves. This is confirmed by Brierley (1934) for *Rubus* sp., and Pirone (1950) for species of Rhododendron, Ilex, Pinus, Picea, and Abies. Roeser (1932) has shown that seedlings of *Pinus ponderosa* and *Pinus contorta* cv. *latifolia* lost thirteen, *Pseudotsuga Douglasii*, eighteen, and *Picea Engelmannii*, twenty-eight times more water at 51°C than at 16°C.

Further substantiation of water loss being responsible for injury was advanced by Brierley and Landon (1946). Latham raspberry has been highly resistant to temperatures as low as minus 45°F, but has been severely injured by minus 8°F, following two days at 37.4°F. They failed to mention the relative humidity or the vapor pressure deficit of the atmosphere for this experiment.

Sunlight may cause excessive heat or an excessively high transpiration rate. Death of trees has occurred as a result of high air temperatures resulting from poor air drainage (Toumey, 1947). He maintained that the amount of injury at any given temperature increased with increasing length of exposure, and the higher the air temperature the shorter the period of exposure required to produce death of plant tissue.

Shirley (1936) made the observation that studies of lethal temperatures for conifers have been confined to the use of seedlings. These results varied considerably, depending on the methods used and the duration of the exposure. Leaves were more exposed to direct sunlight, and were more commonly injured than the other parts of the plant, providing it had developed beyond the juvenile condition.

Kozlowski (1943) found no great difference in the transpiration rate of conifer and deciduous species on a unit area basis.

Screens of canvas, burlap, rustic-woven straw and stakes were suggested by Thompson (1940) to reduce water loss from plants. Foliage and blooms of Gladiolus, Dahlia, Anthirrhinum, and Callistephus were free of sun-scald when grown under cheesecloth enclosures (Batson, 1933).

Wiegand (1906) demonstrated that white paint applied to trunks of peach trees reduced the temperature 15°C.

White, Horsfall, and Talbert (1933) attempted to protect plants by the use of a resin emulsion to which he added short asbestos fibers. Except for the white color, he concluded that the material was of doubtful value. Wax emulsions were used successfully to reduce water loss and to prevent damage to the plants by Miller, Neilson and Bandemer (1937). Gardner (1944) observed that oil-wax emulsion applications reduced water loss 20 to 60 per cent. This reduction depended upon the species, the conditions of the environment, and the concentration of the emulsion.

There were no apparent injurious effects on the foliage when suitable dilutions were used. McMahon in 1947 made a comparison between the use of a wax emulsion and pliofilm cover to reduce the rate and quantity of transpiration of fruit in storage. He concluded that pliofilm was more effective. Sherwood and Hamner (1948) used "Geon 31" to lengthen the life of cut branches of evergreens and some commercial cut flowers. A vinyl resin latex was sprayed on Picea alba (Gartner, O'Rourke and Hamner, 1949). One month after treatment and transplanting without additional water, all unsprayed plants were dead, while 25 per cent of the treated plants survived.

Plastic films developed from high polymers of ethylene are popularly known as "polyethene" or "polyethylene". Sweet (1953) found that moisture was conserved when polyethylene film was used to wrap the rooting medium of cuttings or seedlings.

Hamner, Carlson and Tukey in 1945 reported that succulent plants and cut flowers were maintained in a turgid condition from four to 36 hours longer than control plants by submerging them in water, and evacuating the air by means of a water pump to a pressure of approximately 30 pounds per square inch for 20 minutes. After partial evacuation, the pump was turned off and the pressure gradually increased to that of the air.

R. H. Burris (1952,1953) employed the technique of immersing intact leaves of plants in a solution containing inhibitors of photosynthesis and then evacuating the air from the chamber and the leaves. Normally, he used a vacuum of 15 to 20 mm mercury residual pressure. The leaf was infiltrated by releasing the vacuum, causing the solution to pass into the intercullular spaces of the leaves. The extent of infiltration was evidenced by the change of the leaf from an opaque to a translucent condition. After the leaves were removed, they were blotted dry with paper towels and the petioles immersed in a solution. The excess water was transpired, and the leaves then regained and retained their normal opaque appearance. If the infiltration was too vigorous, the leaves wilted after the infiltrated water was transpired.

Stentor and *Spirogyra*, according to the observations of Greely (1901) exhibited synonymous appearances caused by freezing, desiccation, and plasmolysis.

Studying desiccated *Rubus* canes, Simonds (1942) found that the cell walls were not ruptured, and that sieve tube contents appeared more shrunken than those in turgid tissue. There was a marked deposition of "tannin-like" material, a reduction of size, and a shrinkage of the contents of all living cells exterior to the cambium. He discovered pronounced splitting of the abscission layers. Mix (1916) reported that the youngest xylem cells were the first to be injured, and that complete killing could be identified from discoloration of the cells. According to Parker (1951, 1952) some hypodermal cells of the leaves of *Pinus strobus* had collapsed lengthwise, while the endodermal cells flattened out, and the transfusion tracheids and parenchyma had collapsed from injury. Little change was noted in the xylem and phloem cells. Using excised leaves of *Pinus strobus* and *P. nigra* cv. Austriaca desiccated at room temperature (20^{o}C) and a relative humidity of 60 per cent, Parker (1952) found that the cells of the chlorenchyma, endodermis, and transfusion tissue decreased in size. The adaxial surface of the leaf of the Austrian pine bent inward as contrasted to all three surfaces of the white pine leaves as a result of this shrinkage.

Macroscopic color changes in the leaves may be caused, he postulated, by the contraction of the chlorenchyma. Under the conditions of his experiment, the leaves of *Pinus strobus* dried considerably faster than the *P. nigra* cv. Austriaca.

A breakdown of the nuclear organization of cells of *Prunus* stems subjected to the low temperatures of the winters of 1915-1916 and 1916-1917 was not accompanied by rupture or breaking apart of cell walls (Dorsey and Strausbaugh, 1923). Browning was confined to the cortical cells containing chloroplasts, but injury was of a minor nature. In places where injury was severe, the vessels were occluded by a yellow-brown mass of material which was more pronounced in older vessels.

In his review of literature, Clum (1926a; 1926b) showed that plant tissues in bright sunlight were at a higher temperature than that of the surrounding air, but that there was wide and rapid fluctuations even in periods of short duration of light. In his work only a difference of 2 to 3°C was attributed to transpiration. According to Miller (1938), the actual killing temperature for most plant protoplasm has been found to vary between a high of 113 to 131°F. Lundegårh (1931) observed that there was considerable variation among species in the degree of heat that they are able to withstand. The higher the temperature, the shorter period of time required to produce injury. Sunscald was generally considered by Chandler in 1913 to be the effect of heat absorbed in direct sunlight by the darker colored tissue of the plant. His observations showed that the rate of temperature decrease greatly influenced the killing effect. In late winter the tissues directly exposed to the sun tended to rise above freezing, while those unexposed tissues on the opposite side of a branch remained close to existing atmospheric temperatures. When the sun rays were no longer directly shining on the already warmed periphery of a trunk of a tree, the temperature quickly resumed an equilibrum with that of the surrounding air.

Brooks and Fisher (1926) demonstrated the temperature differential between the exposed and the shaded surface of the fruit of Malus pumila while they were attached to the tree. The exposed side was 10 to 16°F warmer than the shaded side. Black tar-paper coverings on the trunk of fruit trees increased the temperature 10 to 25°F above that of the air (Mix, 1916). Leaves of Nicotiana sp. inserted in cellophane envelopes increased in temperature from 87 to 127°F. Two days later necrotic spots developed. Arthur and Stewart (1933) demonstrated that as the temperature approached the thermal death point from either high radiation or high temperature, the tissue lost great quantities of water.

Potter (1936) concluded that injury incident to cold weather might have been caused by desiccation of the tissues. Allen and Asai (1943) considered the chief problem in growing garden roses to be that of winter injury. They attributed winter injury to be caused by extreme temperature or desiccation of the tissue. Much of the evidence of desiccation as a factor of winter injury has been based on superficial observations. Wilner (1952) found that desiccation occurred mainly during periods with a temperature above freezing, especially above 41°F.

PROCEDURE

One dozen Rosa hybrida cv. Better Times flowers were cut at the Plant Science greenhouse at 1030 hours on May 21, 1953 at various stages of bud development. They were plunged into hot water (98°F). After approximately four hours, the stems were cut to 10 inches in length, and the flowers were separated into comparable groups. Six flowers were placed in a pyrex glass dessicator with a tightly fitted cover. The flowers were above the water, the proximal portion of the stems being held under water by a heavy weight. A vacuum of 20 mm mercury was drawn by means of a pump in order to evacuate the air through a ground glass valve in the cover of the jar. The pressure was equalized gradually (about 5 minutes) to that of the atmosphere. As the vacuum was released, the water passed into the leaves. Vacuum was applied several times until the foliage became water-soaked in appearance. The flowers were removed, allowed to drain on paper toweling, then placed in one-fourth pint jars of water in the laboratory. Four days later, untreated flowers were wilting before petal expansion. The margins of the petals and the leaves were desiccated in contrast to those which had been exposed to the vacuum.

The flowers and foliage of three unidentified cultivars of *Syringa vulgaris* were exposed to a similar treatment. Turgidity was maintained as a result of treatment for 36 hours, an increase of a total of 34 hours over those which were untreated. When flowers were used in the treatment, but failed to be completely submerged in water, they showed no improved keeping quality.

Two one-year-old plants of each of *Syringa vulgaris* cv. Thunberg, *Hydrangea arborescens grandiflora* and *Forsythia spectabilis* were subjected to vacuum treatment. As soon as the cells of the leaves appeared to change color as a result of the entrance of water under reduced pressure, the plants were planted in the garden and no water was added to the soil. Wilting was not discernible on *Forsythia* or *Syringa* during the following week, but diurnal wilting of the succulent growth of *Hydrangea* occurred regularly. Substantial gain in weight was observed as a result of the treatment with vacuum.

Vacuum Treatment of Woody Plants:

One-year-old plants commonly called "lining-out stock" by commercial producers were obtained from Cottage Gardens Nursery of Lansing, Michigan on May 18, 1953. The plants were packed with damp sphagnum surrounding their roots and placed in a controlled temperature of 39^{o}F for eight days. During this period the plants were illuminated, using a low intensity of incandescent lights. The plants were weighed and arbitrary groups selected to contain plants weighing from: 15 to 19 gms, 20 to 24 gms, and over 25 gms. Plants for each treatment were selected so that there were four replications, ten plants per treatment containing the same number of plants from each weight group.

An improved equipment consisting of a pump designed to develop a maximum vacuum of 20 mm of mercury and a tall florists' metal vase of five liter capacity, were used for submerging the plants. This container was placed on a vacuum plate and covered with a pyrex bell jar. During evacuation the plants were weighted to prevent floating.

The roots of the 40 plants were placed in water and allowed to remain during the treatment of the other groups. Forty more plants were subjected to a vacuum for 20 minutes while submerged in tap water, and an additional 40 plants were subjected to the same vacuum treatment using a solution of one ppm indoleacetic acid.

Following treatment, the plants were stored for 12 hours at 39 degrees F before being planted in randomized sequence at the Horticultural Farm of Michigan State College, spaced four feet between rows, and two feet apart within the row. The soil varied from a Hillsdale sandy loam at one end of the row to a silty loam at the other. At the time of transplanting each plant received three-fourths of a pint of water. Initial weights of the plants and weights after treatment, linear measurements and number of branches were recorded at the conclusion of the growing season.

Vacuum Treatment of *Lycopersicum*:

Treatments of *Lycopersicum esculentum* cv. Golden Queen and Red Cherry were identical to those of *Syringa* and *Forsythia*. Roots of seedlings with 4 to 6 true leaves were washed free of soil, plants sorted so that all groups of ten were nearly similar, and treated on June 3, 1953. All plants were transplanted to the field the following day in rows four feet apart with four feet between plants. No supplementary water was supplied at the time of planting or during the growing season. The fruit was harvested at 4-day intervals from August 25 until October 3. The number of fruits and the total weight of fruit of each treatment was maintained.

Soaking Woody Plants:

Plants of *Syringa vulgaris* cv. Thunberg and *Forsythia spectabilis* were soaked for two hours in (a) tap water and (b) one ppm of indoleacetic acid, (c) no soaking. The plants were all transplanted into the field from moist sphagnum packing. Spacing was four feet by two feet apart within the row.

At the conclusion of the growing season, measurements were recorded for the total linear growth and the total number of branches.

Vacuum Treatment of Annual Flowering Plants:

Seeds of the following annual flowering plants were sown in sterilized soil (two parts composted loam, one part sharp sand, one part peat) in flats at the Plant Science greenhouse on December 5, 1953: *Callistephus chinensis* cv. Remo and Kirkwell; *Salvia splendens* cv. St. John's Fire; *Centaurea cyanus* cv. Pinkie; and *Tagetes patula* cv. Dwarf Bedding. The seedlings were treated as in vacuum treatment of woody plants, transplanted January 23, 1954 into 3-inch pots using the same soil mixture as for seeding. Fifteen plants of each cultivar were selected, divided into three groups of five plants each. The number of leaves and leaf areas of two plants were recorded on February 12 and March 6, 1954.

Callistephus was grown at a greenhouse temperature of 50°F with six hours of supplementary incandescent light. The remainder of the plants were grown at 60°F (night temperature).

Polyethylene Covering of Woody Plants:

On February 3, 1954, 40 plants of *Thuja occidentalis* cv. nigra were covered with 0.002 gauge polyethylene film "Cheslene" manufactured by Chester Packaging Products Corporation, Yonkers, New York. One-half of the film was unperforated, and the remainder was perforated with two 6 mm openings per square foot. These plants were transplanted in the nursery. At the time of application the sun was bright and there was a high degree of reflected light from the snow cover on the ground. One month later, thermocouples were attached to the plants under previously applied Cheslene according to the experimental design as shown in Table 1. This was repeated on uncovered plants which were immediately covered in the same manner as before, and an additional pair of thermocouples were placed at 2.5 inch depth in the soil. The thermocouples were attached to a "Minneapolis-Honeywell Brown electronik" continuous temperature recorder. Daily fluctuations of temperature and relative humidity under the various forms of polyethylene protection from March 20 to March 31 was recorded by thirty-hour "Bendix-Friez" hygrothermographs.

One cm stem samples were collected from randomized locations on Thuja occidentalis cv. nigra of both covered and uncovered plants. They were killed in formalin-acetic-alcohol (FAA; 5 cc formalin, 5 cc glacial acetic acid, and 90 cc 70 per cent ethyl alcohol). The tissues were dehydrated using the tertiary-butyl alcohol series, embedded in paraffin, sectioned transversely 8 micra in thickness, and stained with safranin and anilin blue.

Sections were cut from apparently uninjured branches of Thuja occidentalis cv. nigra collected on April 10. The segments used for treatment were seven cm from the apex of the branches. One-half of the material was stored in tap water and the other half were left dry. Three stems of each of these treatments were subjected to the following time and temperature series (see Table 2).

TABLE 1

The Location of Thermocouples at Each Tree Used in the Course of These Studies

Thermocouple No.	Where Installed on Tree, Type and Date of Cover	Position in Respect to Tree
1	Middle of plant, perforated cover 2-3-54	South
3	Apex, perforated cover 3-6-54	South
5	Apex, non-perforated cover 2-3-54	South
6	Apex, non-perforated cover 3-6-54	South-west
7	Middle, perforated cover 3-6-54	East
8	Apex, perforated cover 2-3-54	South-west
9	Apex, not covered	Tip
10	Middle, non-perforated cover 3-6-54	West
11	Middle, not covered	North
12	Open soil, 2 1/2 inches Deep	None
14	Middle, non-perforated cover 2-3-54	South
16	Soil under Tree, 2 1/2 inches deep	Southwest

TABLE 2

Time and Temperature Treatment, _Thuja occidentalis_ cv. Nigra Samples

Treatment	Length of Time of Treatment in Minutes			
	15	30	60	120
	Actual Temperature in Degrees F			
No water	140.0	140.0	140.0	140.0
In water	105.0	110.0	125.6	136.4
No water	104.0	104.0	104.0	104.0
In water	86.0	95.0	100.4	107.6
No water	32.0	32.0	32.0	32.0
In water	50.0	44.0	41.0	---
Room temperature				
No water	75.0	75.0	75.0	75.0

RESULTS

Vacuum Treatment of Woody Plants:

The average total length of all stems of *Syringa vulgaris* cv. Thunberg were not significantly influenced by vacuum treatments of water or indoleacetic acid (Table 3). Growth of stems was somewhat inhibited by the water treatment, but more so by the addition of the indoleacetic acid (Table 3).

All plants subjected to a vacuum increased the number of stems produced from axillary buds. Plants treated with water produced an average of 12, whereas those receiving indoleacetic acid treatment produced 11.2 branches. There seemed to be suppression of growth of lateral branches, but with an increase in number of branches (Table 3).

Forsythia spectabilis, on the contrary, showed a suppression in the average linear growth by treatment with a vacuum. Stem elongation in both treated groups was significantly less when compared to plants given no vacuum treatment. Table 3 shows that indoleacetic acid had a decided depressing action on the linear growth of lateral branches.

The average number of branches (Table 3) was likewise less when the plants were treated with water or when

TABLE 3

Length and Amount of Growth as Influenced by Vacuum Treatment (40 Plants per Treatment)

	Untreated	Water	Indoleacetic Acid
Syringa vulgaris			
Average number of stems	10.0	12.0	11.2
L.S.D. 5% 1.39			
L.S.D. 1% 2.11			
Average length of stems (cms)	107.1	98.1	95.8
L.S.D. 5% 20.6			
L.S.D. 1% 31.2			
Forsythia spectabilis			
Average number of stems	43.6	37.3	34.1
L.S.D. 5% 3.8			
L.S.D. 1% 5.8			
Average length of stems (cms)	1545.4	1211.2	1040.6
L.S.D. 5% 129.6			
L.S.D. 1% 196.3			

indoleacetic acid was added.

In descending order, significantly less branches were produced on plants receiving the following treatments: untreated, water under vacuum, and indoleacetic acid under vacuum.

Vacuum Treatment of Lycopersicum:

The total number of fruits produced by the indoleacetic acid treated cultivar Red Cherry was not significantly different at the 5 per cent level than the number produced by the untreated plants. However, the indoleacetic acid treated plants did produce a significantly greater number of fruits than those subjected to a vacuum treatment of water (Table 4). The total weight of fruit produced on ten plants from the water treatment was significantly less than either the untreated or the indoleacetic acid treated plants. No statistical differences were found between the yield of the untreated and indoleacetic acid treated plants.

The cultivar Golden Queen did not respond to treatment in the manner of cultivar Red Cherry. There was no significant difference in the total number of fruits or the weight of the fruit from treated and untreated plants. It may be noted in Table 4 that the water treated plants produced the lowest number

TABLE 4

Yield of Fruit of Lycopersicum esculentum cvs. Red Cherry and Golden Queen (10 Plants per Treatment)

	Treatment		
	Untreated	Water Under Vacuum	Indoleacetic Acid Under Vacuum
		Number of Fruit	
Cultivar			
Red Cherry	918	693	976
L.S.D. 5% 248.4			
L.S.D. 1% 456.1			
Golden Queen (Total)	1599	1258	1439
L.S.D. 5% 728.9			
L.S.D. 1% 1338.5			
Golden Queen (Early)	155	166	133
L.S.D. 5% 89.4			
L.S.D. 1% 164.1			

of fruit and the least total weight. The untreated plants produced the most tomatoes. The early yields (total harvest for first two-week period) of all treatments were similar in the number and total weight of fruits harvested (Table 5). Observations in the field indicated that the number of fruit on each cluster was greater from the indoleacetic acid treated plants. This was not shown to be of statistical significance.

TABLE 5

Weight of Fruit - Lycopersicum esculentum cvs. Red Cherry and Golden Queen (10 Plants per Treatment)

	Treatments		
	Untreated	Water Under Vacuum	Indoleacetic Acid Under Vacuum
	Weight of Fruit (Pounds)		
Cultivar			
Red Cherry	22.6	15.4	23.3
L.S.D. 5% 6.3			
L.S.D. 1% 11.6			
Golden Queen (Total)	793.9	650.3	668.3
L.S.D. 5% 301.1			
L.S.D. 1% 553.0			
Golden Queen (Early)	97.5	99.0	80.4
L.S.D. 5% 51.2			
L.S.D. 1% 94.0			

Soaking Woody Plants:

Plants of Syringa showed an increase in the average total linear growth as a result of a soaking treatment in water. Those plants whose roots had been soaked gained an average of 9.2 cm per plant, but linear growth was depressed by the addition of indoleacetic acid to the soaking solution.

The average number of lateral branches produced on each plant was significantly greater on the treated plants, however a greater number was produced on those plants the roots of which had been soaked in water for two hours (Table 6). One ppm indoleacetic acid appeared to depress growth as measured by the average total length of new growth of *Forsythia*. There was an indication that soaking the roots of *Forsythia* in water would increase the average total length of all above ground parts.

Vacuum Treatment of Annual Flowering Plants:

From Tables 7 and 8 it is apparent that there was little consistency in the growth of plants subjected to the various treatments. *Salvia splendens* cv. St. John's Fire had a greater leaf area on the two sampling dates. By the later date, *Salvia*, *Callistephus* cv. Kirkwell and *Centaurea*, as a result of treatment with indoleacetic acid had a greater leaf area than the untreated or the water treated plants.

There was an increase in the number of lateral branches and leaves of *Salvia* plants which had undergone treatment.

In general, there were very slight differences in the number of leaves produced between the treated and untreated plants. Genera as well as cultivars responded differently to treatment with water and indoleacetic acid.

TABLE 6

Influence of Soaking on Growth (Average of 40 Plants per Treatment)

	Treatment		
	Untreated	Soaked in Water	Soaked in Indole-acetic Acid
Syringa vulgaris			
Average number of stems	13.4	15.1	14.3
L.S.D. 5% 0.9 L.S.D. 1% 1.3			
Average length of stems (cms)	79.9	89.1	76.1
L.S.D. 5% 7.2 L.S.D. 1% 10.4			
Forsythia spectabilis			
Average number of stems	36.2	37.3	38.4
L.S.D. 5% 2.70 L.S.D. 1% 3.93			
Average length of stems (cms)	1126.4	1170.0	1008.2
L.S.D. 5% 55.0 L.S.D. 1% 80.0			

TABLE 7

Leaf Area (Sq. Cms.) as Influenced by Vacuum Treatments (2 Plants per Treatment)

Date	Genera	Treatment		
		Untreated	Water	Indoleacetic Acid
February 17	Salvia	6036	6796	9444
	Callistephus cv. Remo	4868	4984	6624
	Callistephus cv. Kirkwell	5260	7404	4796
	Centaurea	7040	9074	7352
March 6	Salvia	75016	69068	87860
	Callistephus cv. Remo	17588	10592	15388
	Callistephus cv. Kirkwell	16528	19608	22792
	Centaurea	15820	18096	18440

TABLE 8

Number of Leaves and Stems as Influenced by Vacuum Treatments (2 Plants per Treatment)

Date	Genus	Treatment					
		Untreated		Water		Indoleacetic Acid	
		Leaves	Branches	Leaves	Branches	Leaves	Branches
February 17	Salvia	8	0	10	0	12	0
	Callistephus cv. Remo	11	0	10	0	10	0
	Callistephus cv. Kirkwell	7	0	10	0	7	0
	Centaurea	8	0	9	0	9	0
March 6	Salvia	68	13	78	16	76	16
	Callistephus cv. Remo	26	0	22	0	22	0
	Callistephus cv. Kirkwell	15	0	19	0	19	0
	Centaurea	14	0	17	0	16	0

<u>Polyethylene Covering on *Thuja occidentalis*:</u>

Air temperature as well as temperatures at various locations on plants of *Thuja occidentalis* cv. nigra fluctuated widely during a 25-day period in March 1954. Temperatures on unprotected plants were consistently lower than existing official temperatures recorded by the Weather Bureau in the same vicinity. Temperatures of plants covered with "Cheslene" protection were higher. The temperatures under perforated film were slightly lower (1 to 3°F) than under non-perforated cover. The highest temperatures were observed between 1200 and 1500 hours, and the lowest temperatures were at 0600 hours. The maximum soil temperature was 50°F with diurnal fluctuations of approximately 28°F.

For 17 days during the experiment snow was on the ground. Total sunshine during the month of March was 204 hours, only 55 per cent of the maximum.

Desiccation and browning was prevalent on all covered trees, which was more intense on the south and southwest exposure and concentrated at the apex. This injury did not always originate at the apex of the branch, nor was injury limited to points of contact between branches and the plastic. Injury was of two types (a) desiccation and browning of the leaves and branches;

and (b) desiccation without the accompanying browning.

Desiccation occurred first on the exposed side of the branch, and gradually affected the entire branch. At later stages, the branch was brittle. Frequently, desiccation was evident and browning did not develop.

Relative humidity fluctuated between 100 per cent during the periods of lowest temperature, frequently decreasing to 20 per cent during periods of higher temperatures. Evaporation was reduced by the polyethylene cover because of low moisture movement capacity of the film, and the reduced air movement within the enclosure. Water frequently condensed in the folds of the plastic with a lowering of the temperature.

Temperature-relative humidity relationships of air, under non-perforated and under perforated transparent polyethylene are shown in Figures 1, 2 and 3.

The plastic film was subject to rapid breakdown by splitting under field conditions during March 1954.

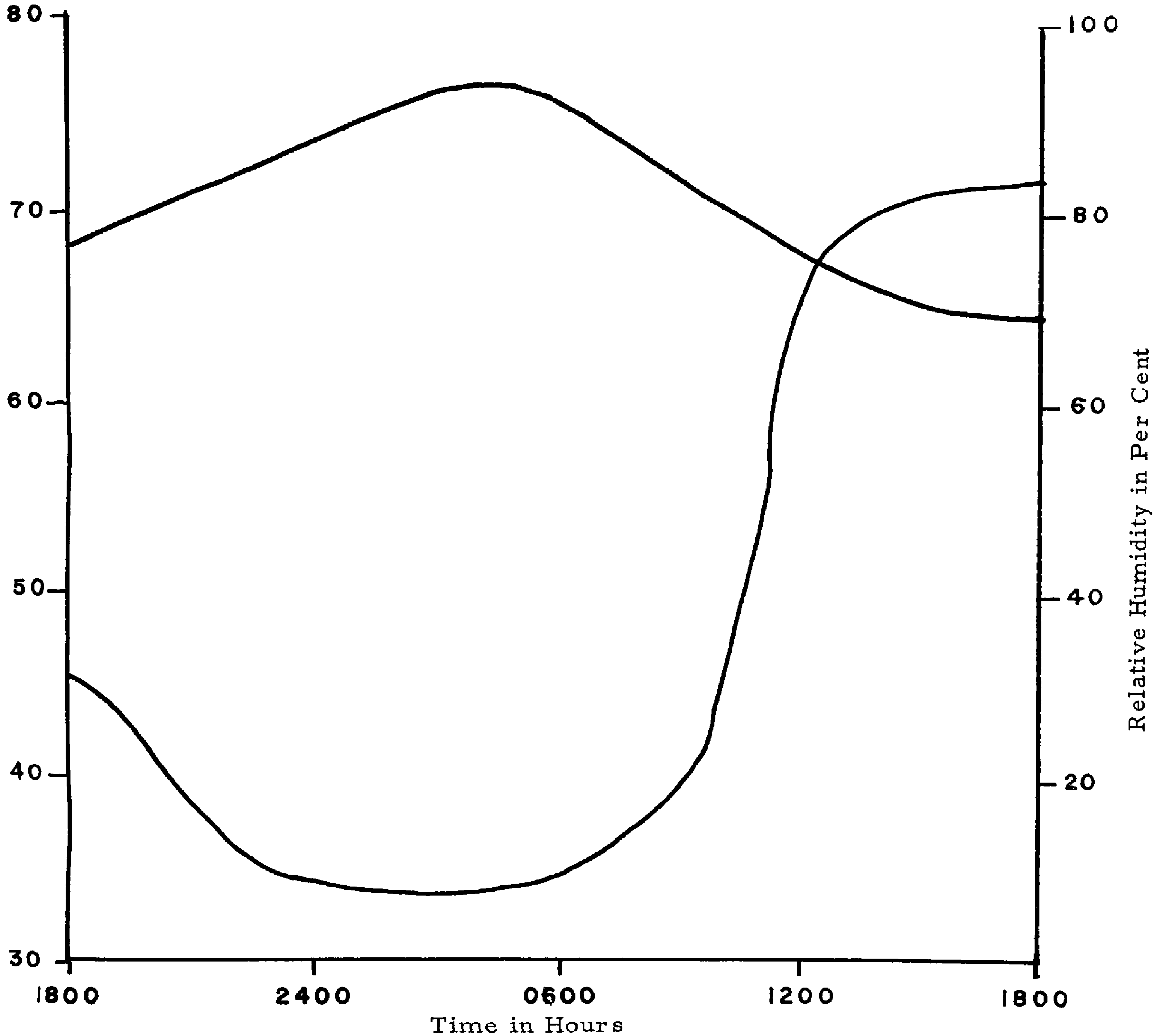

Figure 1. Temperature and relative humidity of open air over representative 24 hour period.

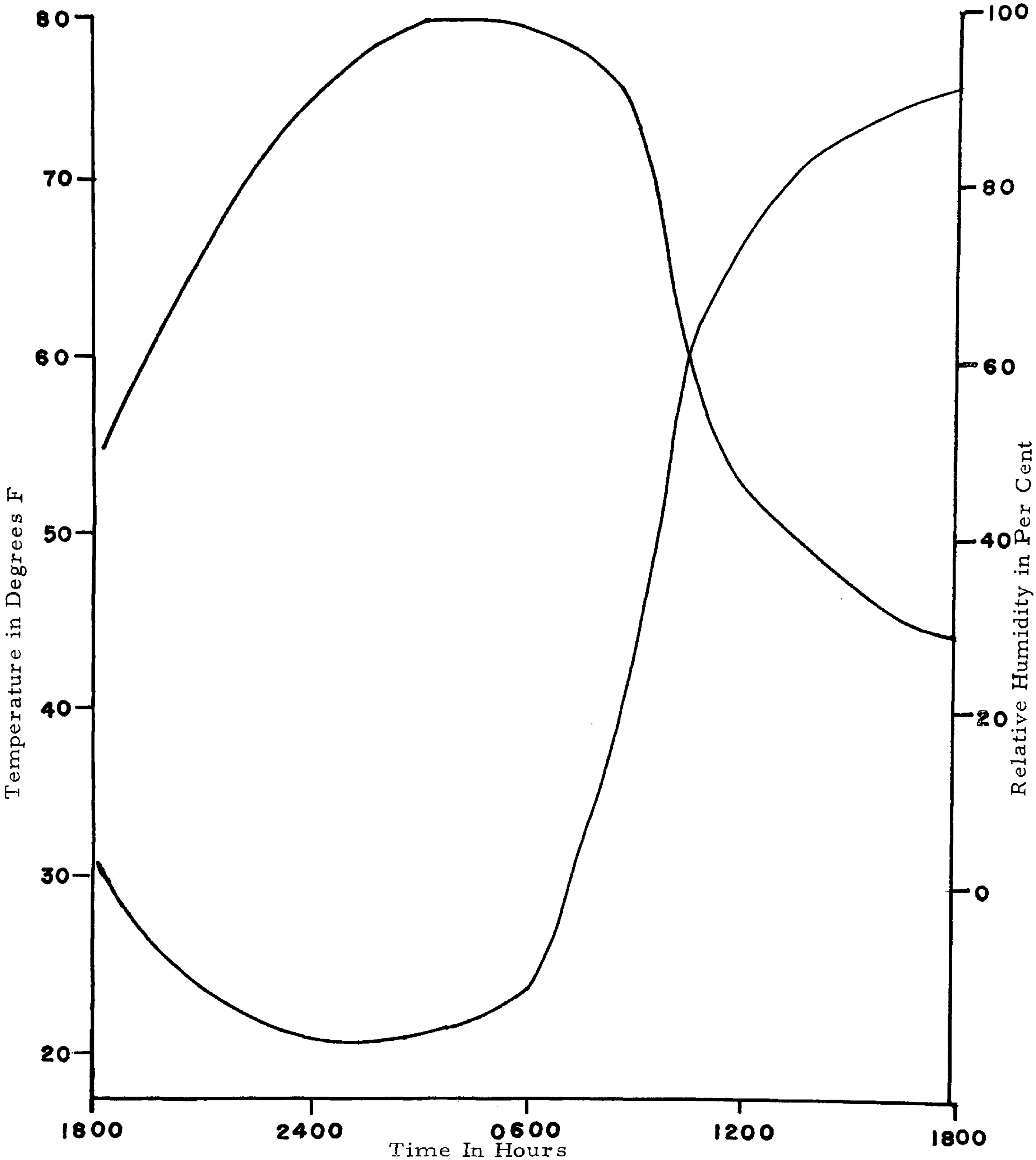

Figure 2. Four day average of temperature and relative humidity for air enclosed in a perforated polyethylene cover.

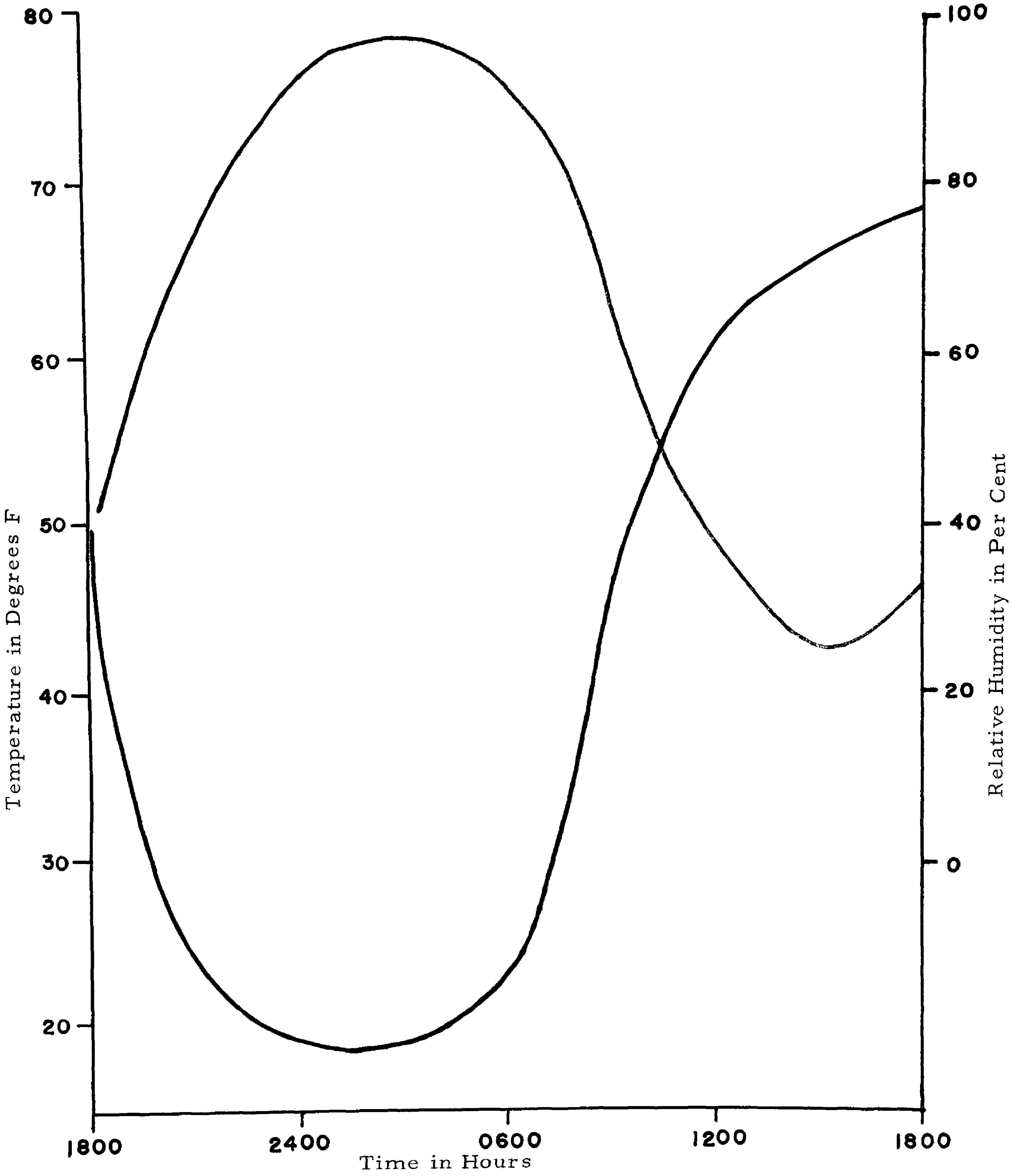

Figure 3. Four day average of temperature and relative humidity for air enclosed in non-perforated polyethylene cover.

Tranverse sections of six pairs of scale-like leaves were illustrative of the decussate phyllotaxy of *Thuja occidentalis* cv. nigra (Figure 4). The central cylinder was in early stages of unlignified, but thickened primary xylem differentiation. Most of the cells were irregular parenchyma with dense cytoplasm and prominent nuclei.

The individual leaves were protected by a heavy somewhat scalloped cuticular covering on the abaxial surface (Figure 5). Frequent deposits of unidentified materials appeared to be buried in the cuticle. This cuticle was approximately twice the depth of the subtending epidermis, to which it appeared to be connected by plasmadesmata.

The epidermis consisted of a single row of tangentially elongated cells, with spine-like projections on the abaxial surface. The walls of the epidermis were of cellulosic composition, thick, with conspicuous cytoplasm and nuclei. Stomata were not frequent in their distribution over the surface.

Directly beneath the epidermis was a single or double layered hypodermis composed of collenchyma cells which were polygonal in transverse section. The cell walls were extremely thick, penetrated by simple pits and contained very small lumen.

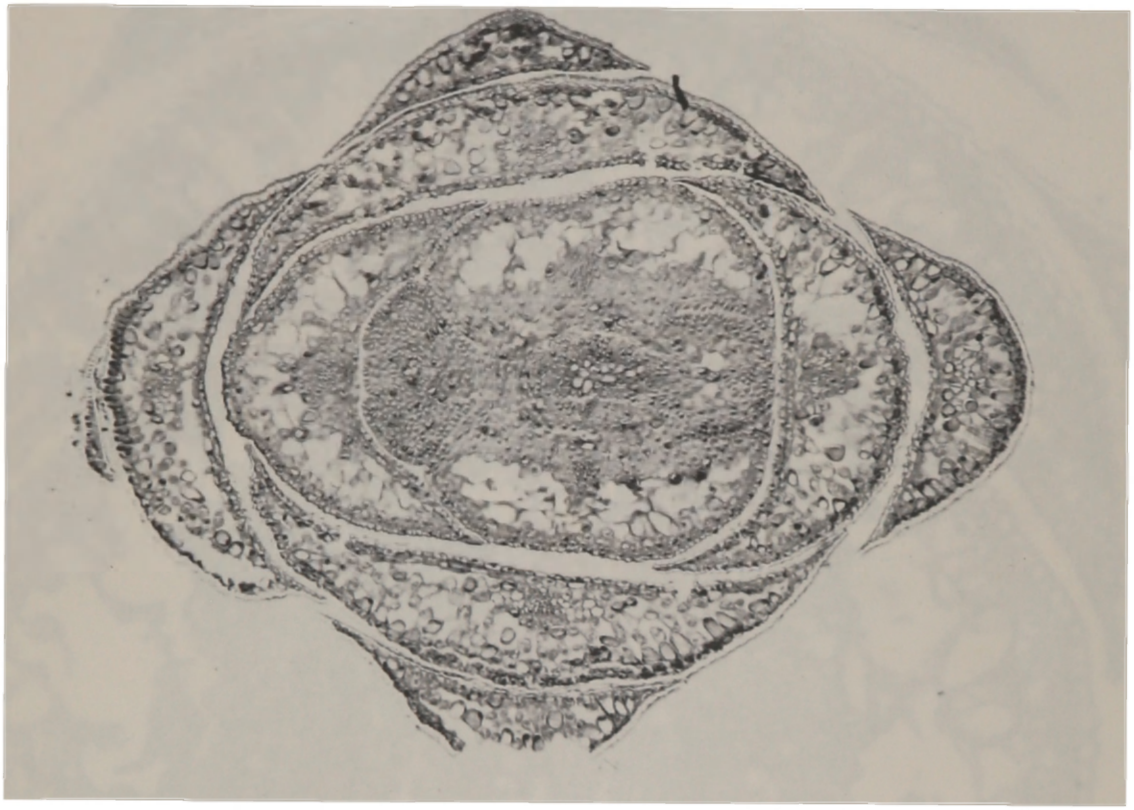

Figure 4. Transverse section of stem and leaves of Thuja occidentalis cv. nigra, one cm from the apex (X80).

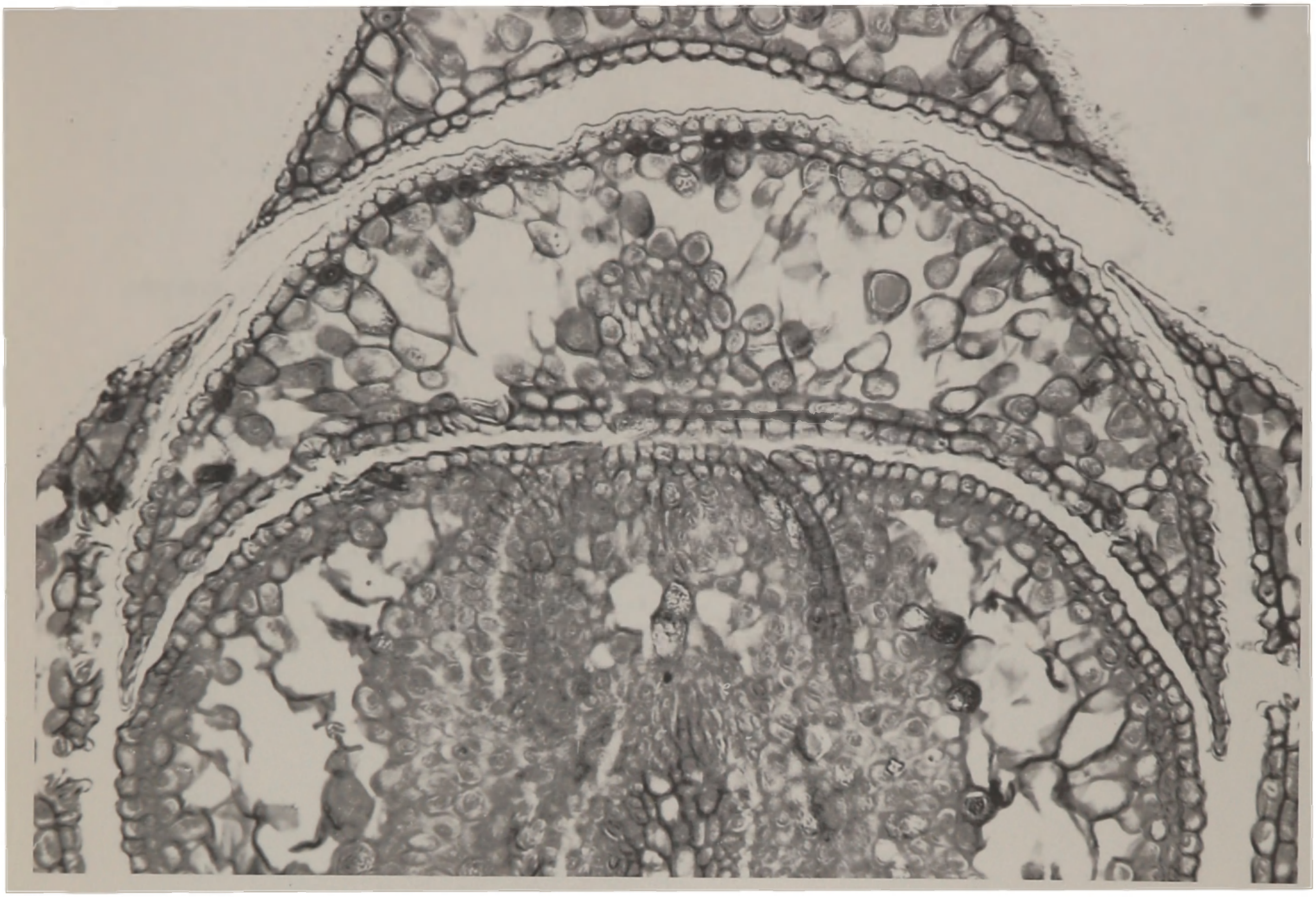

Figure 5. Transverse section of one complete leaf of Thuja occidentalis cv. nigra, one cm from the apex of the stem (X240).

A single layer of elongated palisade-like parenchyma cells was located beneath the hypodermis. These cells were irregular in size and arrangement, and contained dense cytoplasm with large prominent nuclei. Surrounding the specialized tissues were irregularly shaped parenchyma cells, loosely attached with large intercellular air spaces. Conspicuous deposits of tannin were found in the parenchymatous mass of tissue.

The vein was surrounded by a layer of heavy-walled endodermal cells which were somewhat irregular in shape and usually radially flattened. Surrounding the adaxial xylem and abaxial phloem were irregularly shaped parenchymatous and tracheid cells termed transfusion tissue.

Adjacent to the adaxial epidermis was a single layer of tangentially elongated parenchyma. The epidermis adjacent to the centrally located stem was composed of large cells with convex outer walls, often interrupted by sunken stomata (Figure 6) with their subsidiary cells arching over the guard cells. A portion of the guard cell walls were lignified and partly cellulosic. This epidermis is covered by a thin layer of cuticle, except over the subsidiary cells where heavier deposits were found.

Adjacent to the abaxial surface of mature leaves one or more resin ducts were found, which were lined by elongated epithelial cells (Figures 7 and 8).

In older stems, the pith cells were surrounded by extremely heavy cellulose walls (Figure 9). The ray cells of the xylem were greatly elongated toward the periphery of the stem. They were parenchymatous, arranged in a uniseriate row, becoming more tear-drop shaped cells in the phloem region. The cambial zone separated the xylem cylinder and the phloem, which was composed of alternating layers of sieve cells, fusiform and storage parenchyma. The primary phloem and cortex were an indistinguishable crushed mass.

Naturally occurring injury on Thuja occidentalis cv. nigra, although difficult to identify, generally manifested itself as a loss of the dark green color characteristic of the cultivar. On the parts exposed to the continuous south-west winds, this plant became reddish-olive green near the apices of the branches. This condition was difficult to distinguish from injury which was usually termed winter injury, because the color was believed to be a naturally occuring phenomenon

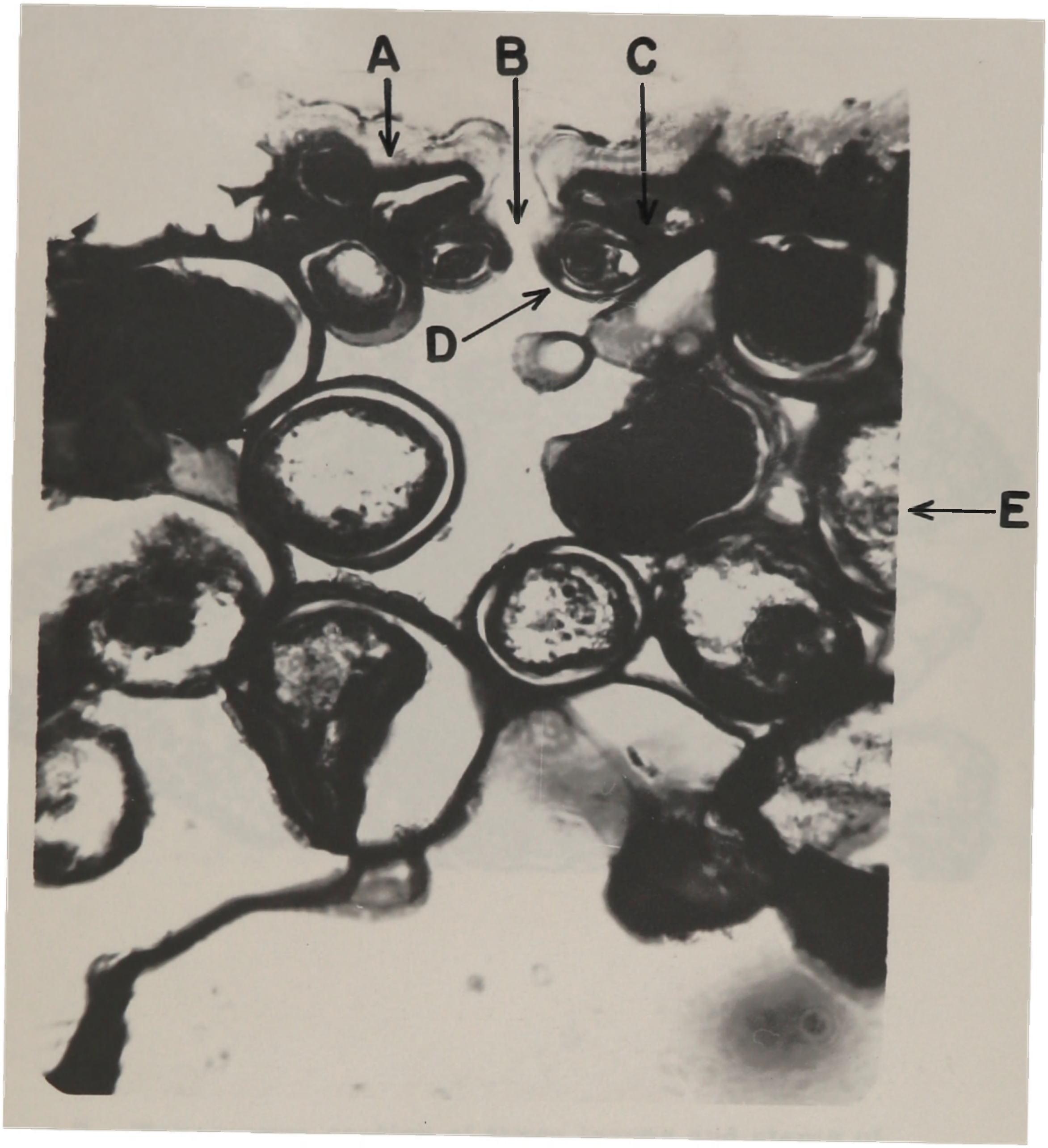

Figure 6. Transverse section of outer layers of cells of leaf of Thuja occidentalis cv. nigra showing the cuticle A, stoma B, subsidiary cells C, guard cell D, and adjacent mesophyll E (X1000).

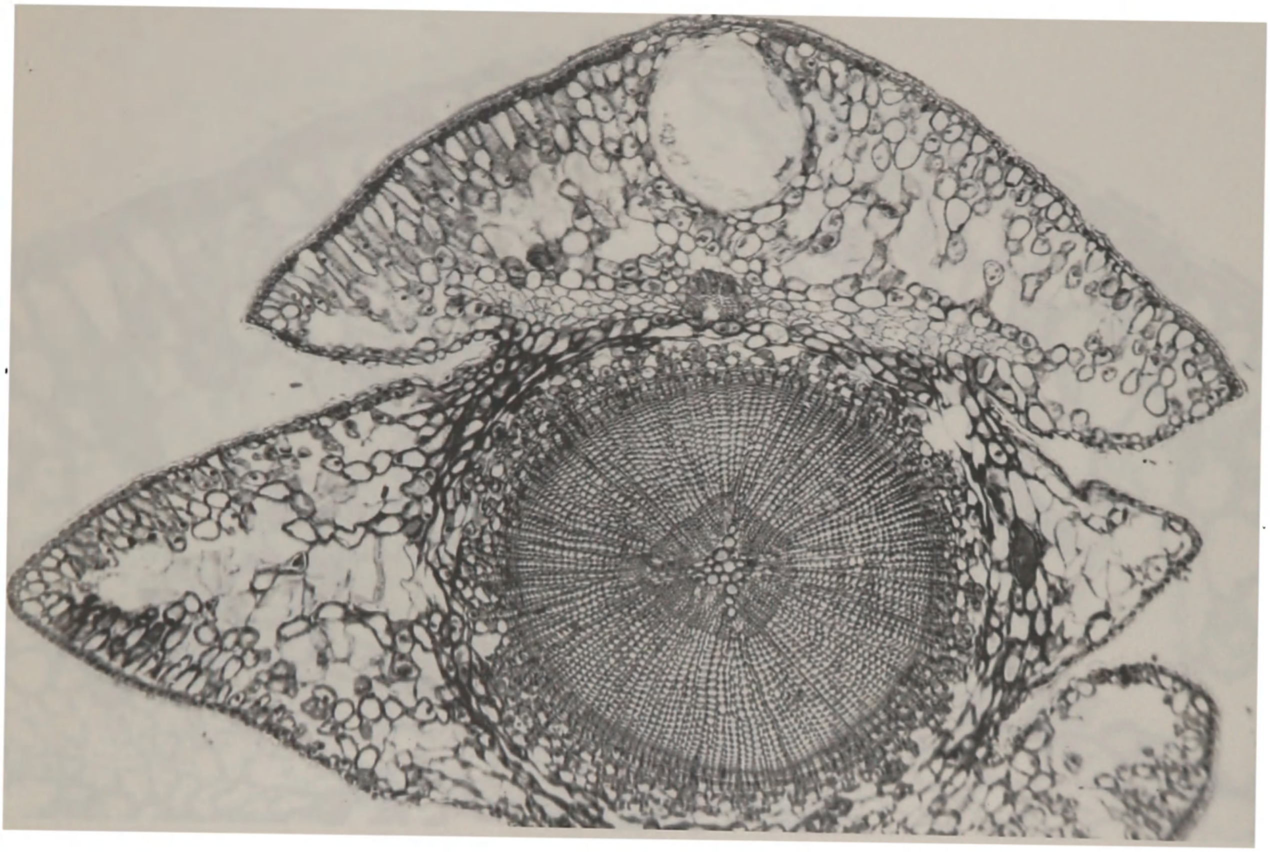

Figure 7. Transverse section of three leaves and stems of Thuja occidentalis cv. nigra six cms from the apex showing a large resin duct in the mesophyll (X80).

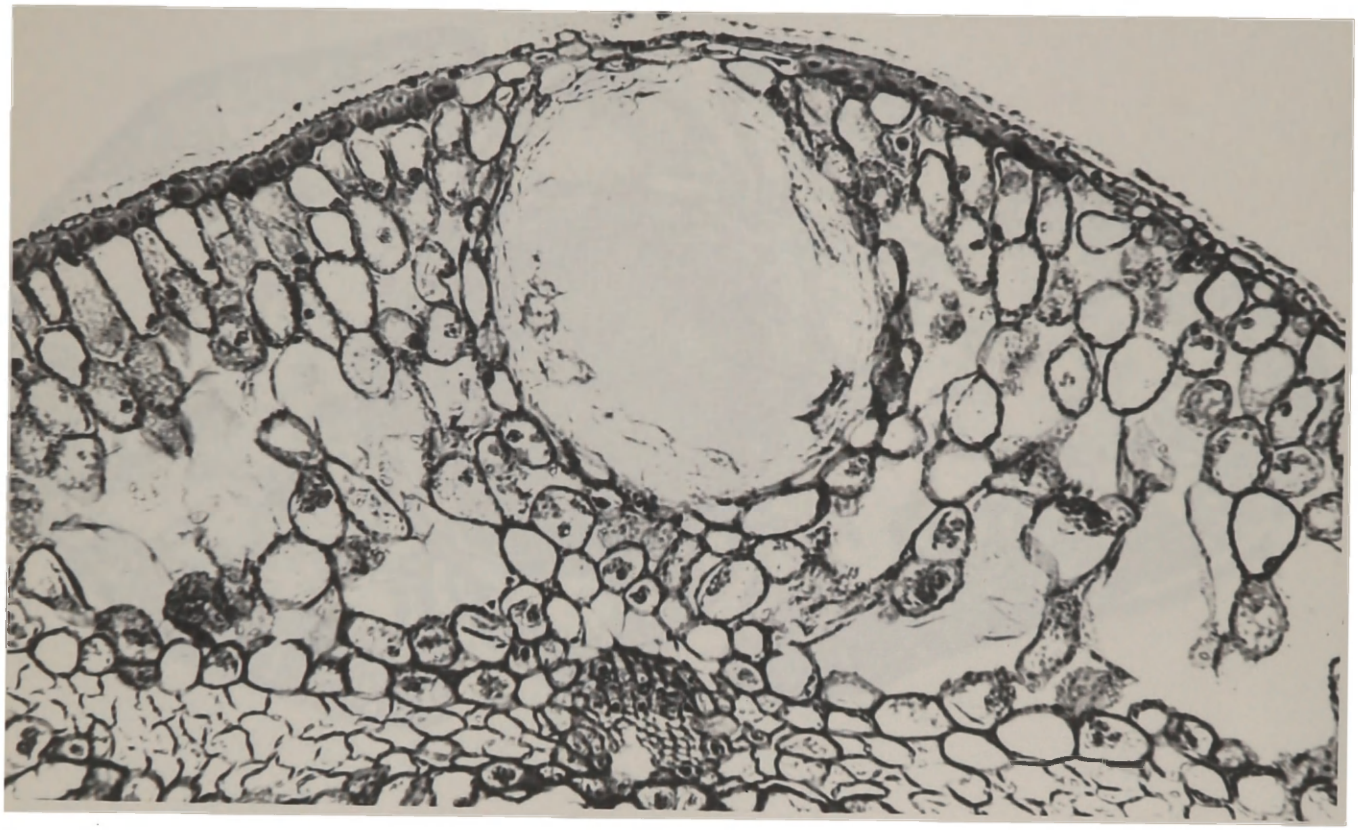

Figure 8. Transverse section of a leaf of Thuja occidentalis cv. nigra six cms from the apex showing a large resin duct in the mesophyll (X240).

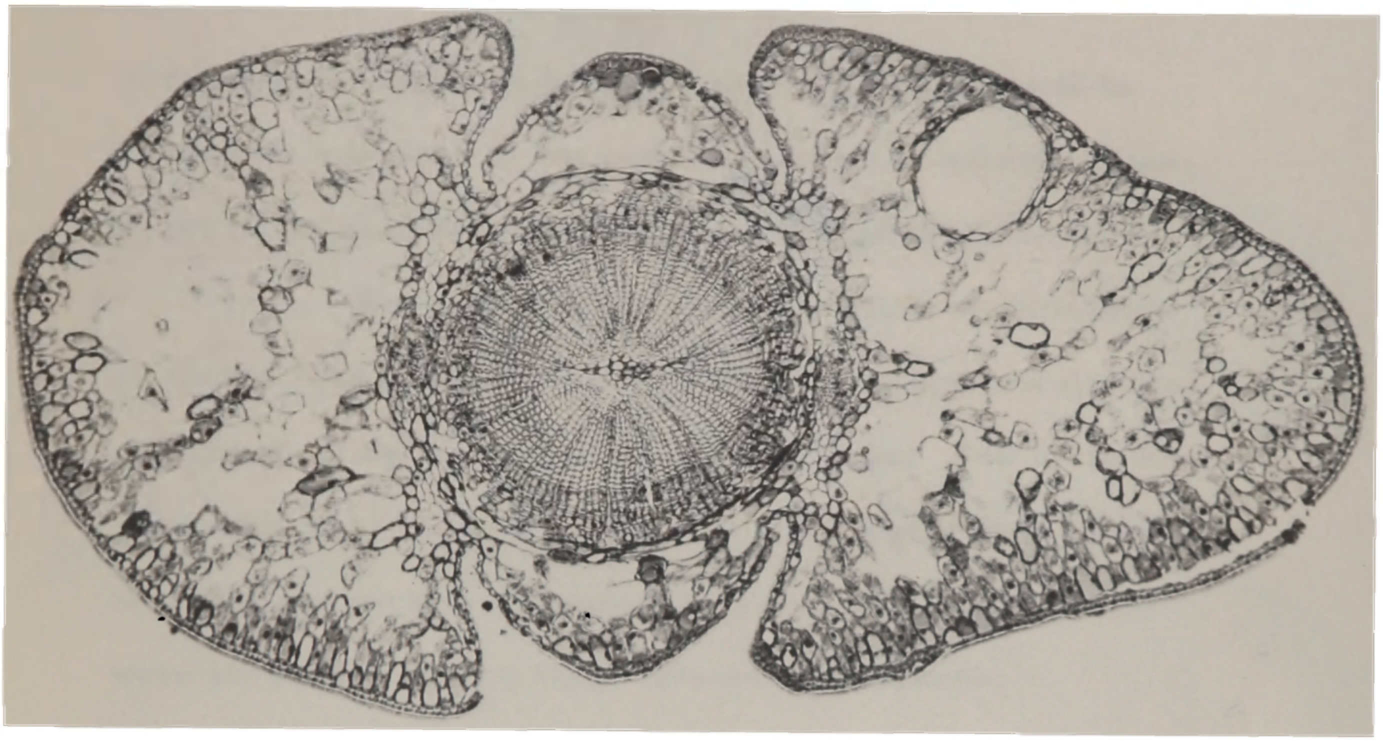

Figure 9. Transverse section of mature leaves and stem of Thuja occidentalis cv. nigra six cms from apex (X80).

of the genus. Injury appeared to originate on the south to south-west exposure of the tree. This type of injury is illustrated in the accompanying photomicrograph (Figure 10).

The most conspicuous discernible modification of the leaf was the inactivation of the abaxial epidermal cells, brought about by the deposition of a mucilage. Few cells were somewhat plasmolyzed. Epidermal and hypodermal cells contained lumen packed with tannin-like deposits. These cells were shrunken, leaving large intercellular spaces.

Palisade-like cells were found to be shrunken in different patterns (Figure 11). In general, the walls of the longest dimension became concave, accompanied by a shrinkage of the protoplasm (Figure 12). Intercellular spaces were prevalent and relatively large. Cells of mesophyll were irregularly arranged, with the varying conditions of protoplasm shrinkage being of paramount interest. In some cases the cytoplasm appeared to be almost completely absent. The phloem elements of the vascular strand were irregular in shape and condition. The cytoplasm was extremely dense, and some cells appeared physiologically inactive.

The xylem cells seemed to be little affected. The

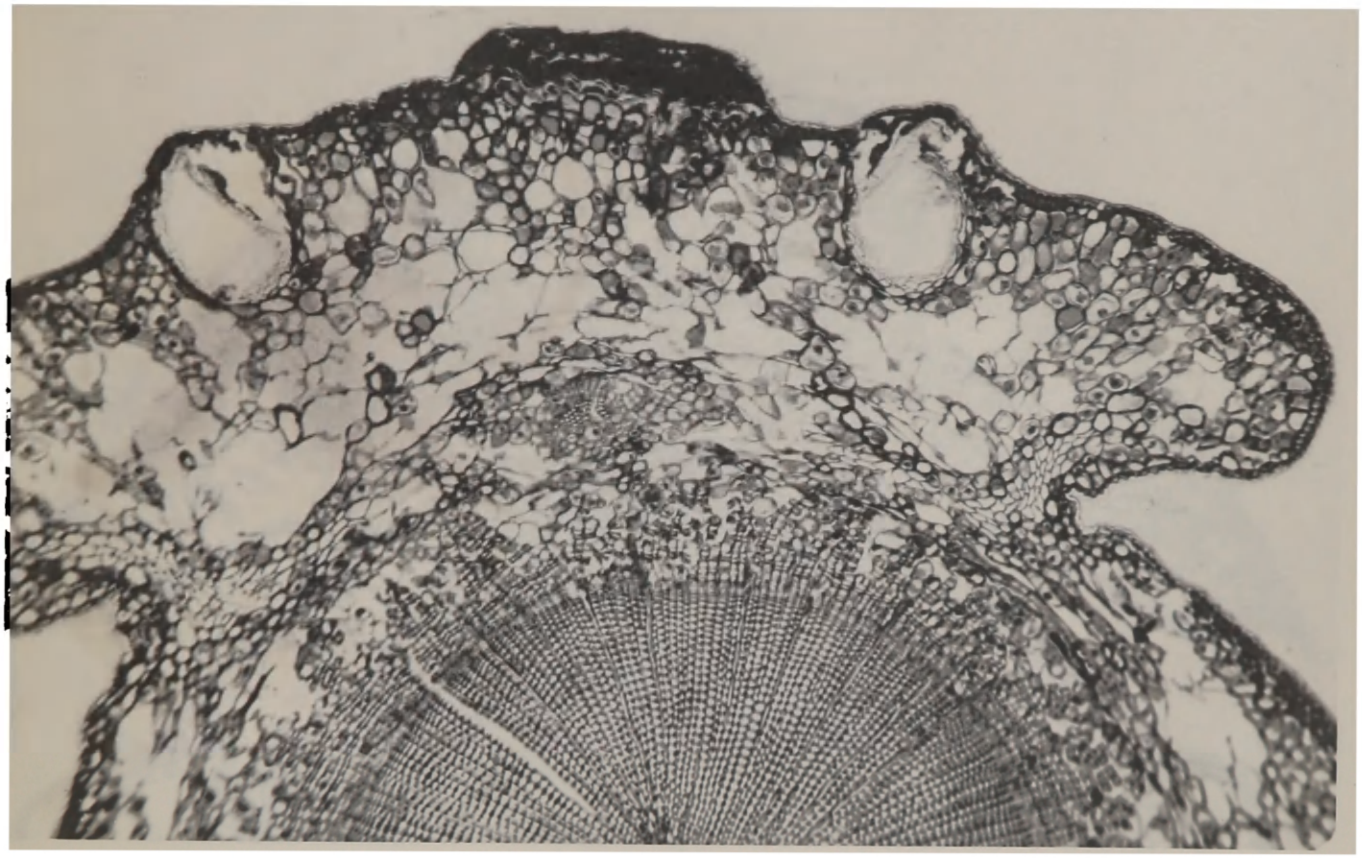

Figure 10. Transverse section of part of leaf and stem of Thuja occidentalis cv. nigra ten cms from the apex showing the injury developing on the outer edge of the exposed leaf (X80).

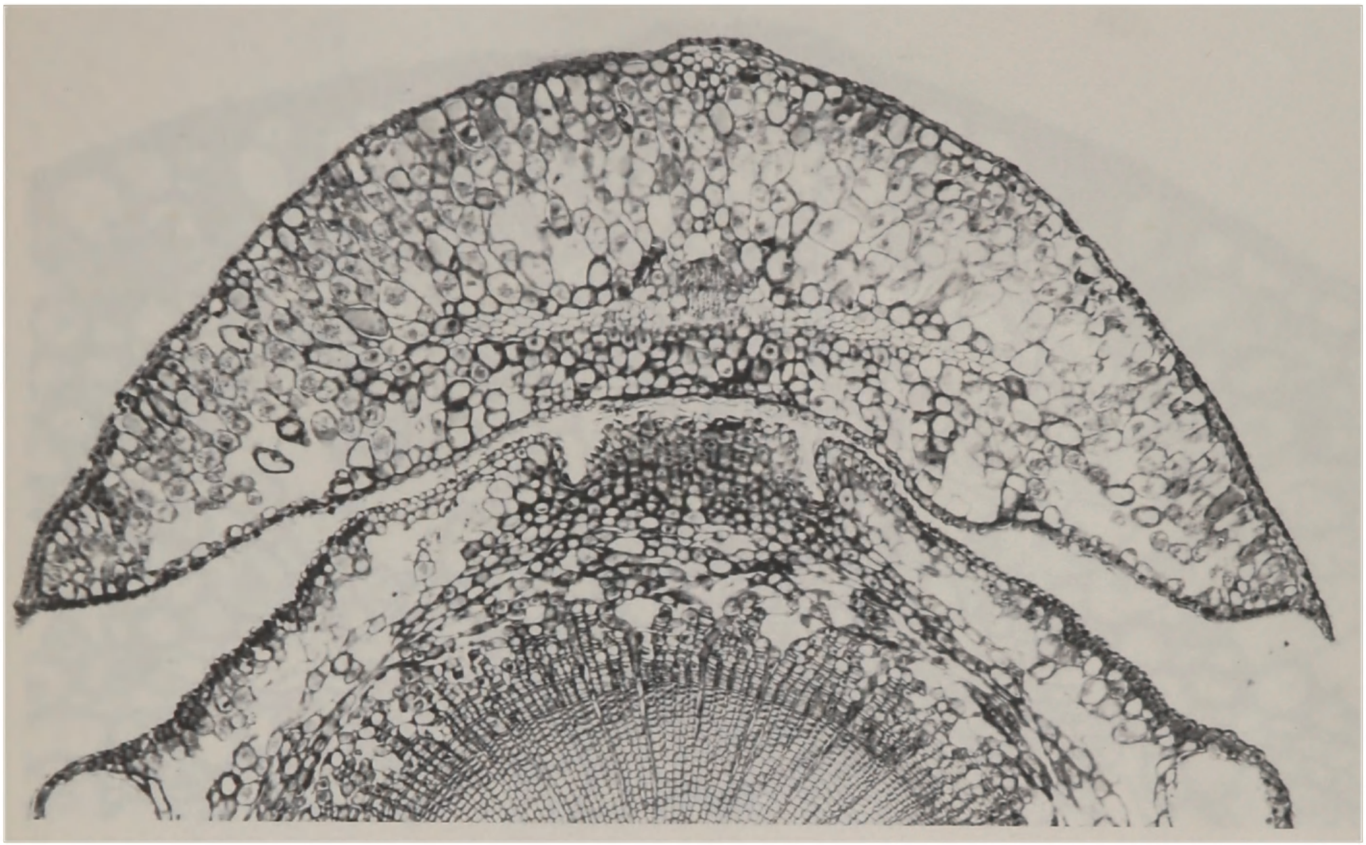

Figure 11. Transverse section of a leaf and part of a stem of <u>Thuja</u> <u>occidentalis</u> cv. nigra eight cms from the apex showing initial stages of dehydration on plants in the open field (X80).

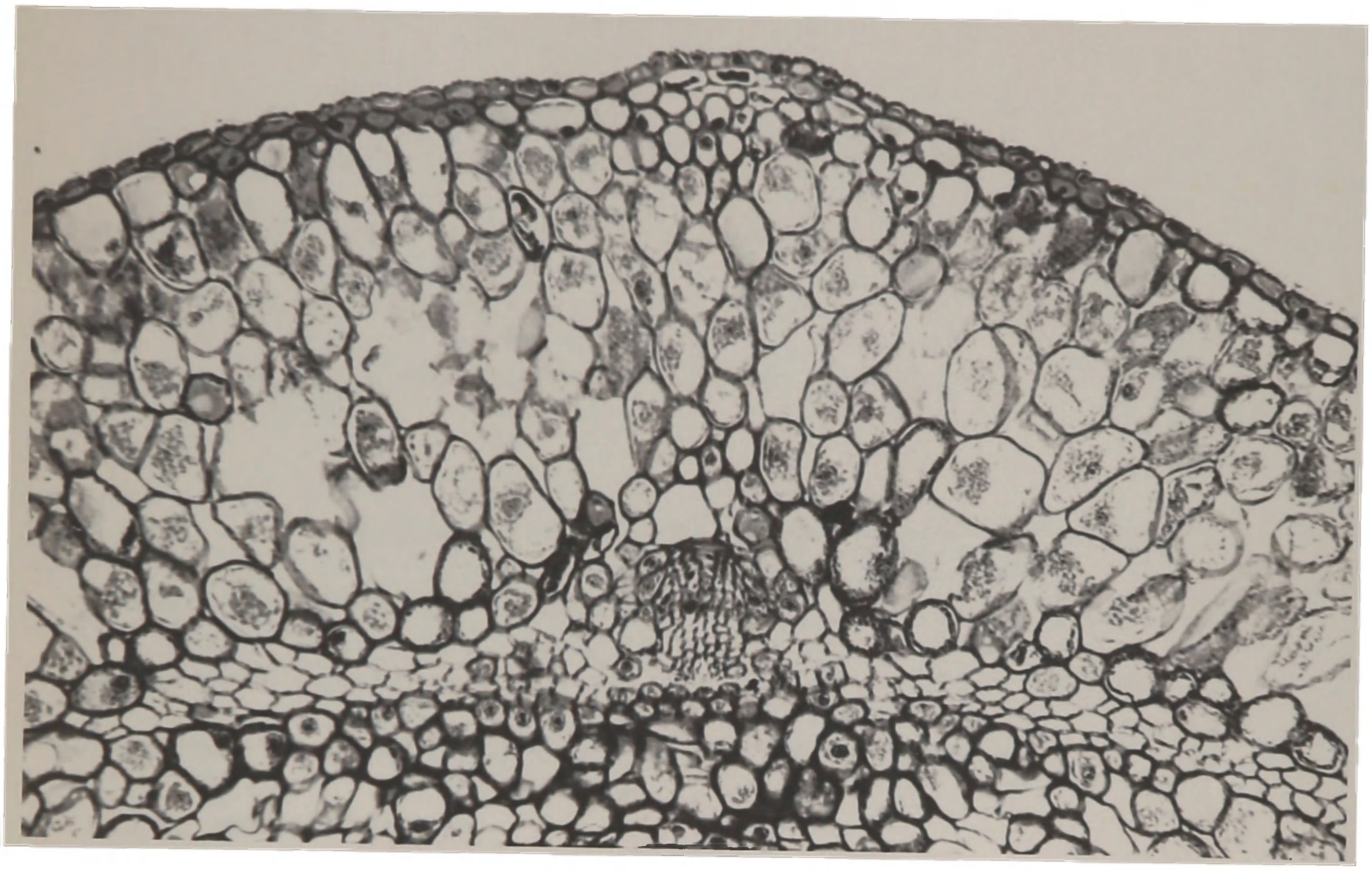

Figure 12. Transverse section of part of leaf of Thuja occidentalis cv. nigra eight cms from the apex showing initial stages of dehydration on plants in the open field (X240).

transfusion tissue surrounding the vascular strand was collapsed, giving the appearance of radial stretching. Slight modifications of the endodermal cells were principally wrought by shrinkages of the cytoplasm and the cell walls to a lesser extent.

The adaxial epidermal cells appeared to be functional, but with minor depositions of the tannin materials. The cells were somewhat flattened. No rupturing of cell walls was observed in any of the preparations. The cuticle remained thick, and unchanged.

Injury to the leaves of plants covered with perforated and non-perforated polyethylene was in the form of chlorophyll destruction. The leaf was first pale green, yellow and finally brown. The chloroplasts of the leaves of *Thuja occidentalis* cv. nigra were distributed in the parenchyma of the mesophyll. Cellular contents of the abaxial epidermis were atrophied. Dense tannin materials filled the lumens of most of the epidermal and collenchyma cells. All cells of the mesophyll layer were contracted, with a greater degree of plasmolysis and presence of deposition products within the cell.

Severe injury which ultimately caused the death of the plant produced extreme atrophy of the epidermal cells, the abaxial surface of which was pronouncedly convex (Figure 13). Contents of all cells were extremely reduced in area, with a marked derangement of the cells. Rupturing of cell walls was exceedingly rare, even at the protoplasmic death point. In the stem, all parenchymatous cells, regardless of location, were shrunken, contained deposits of tannins, and were deformed. Injury to the xylem cells and fibers was not consequential. Parenchyma in the pith region was also affected, but usually without rupturing of the cell walls.

In leaf and stem parts of _Thuja_ _occidentalis_ cv. nigra, which had been artifically frozen, the cuticle, epidermis, and hypodermis were not observed to differ from uninjured tissues (Figure 14). The location of the parenchyma cells of the mesophyll was greatly altered by freezing. Although some plasmolysis appeared, the principal injury was the rupture of the cell walls of many cells, both in the leaves and in the stem. Extensive rupturing occurred at the union of pith and primary xylem, in some cases following the xylem ray toward the periphery of the stem. Xylem cells were somewhat separated.

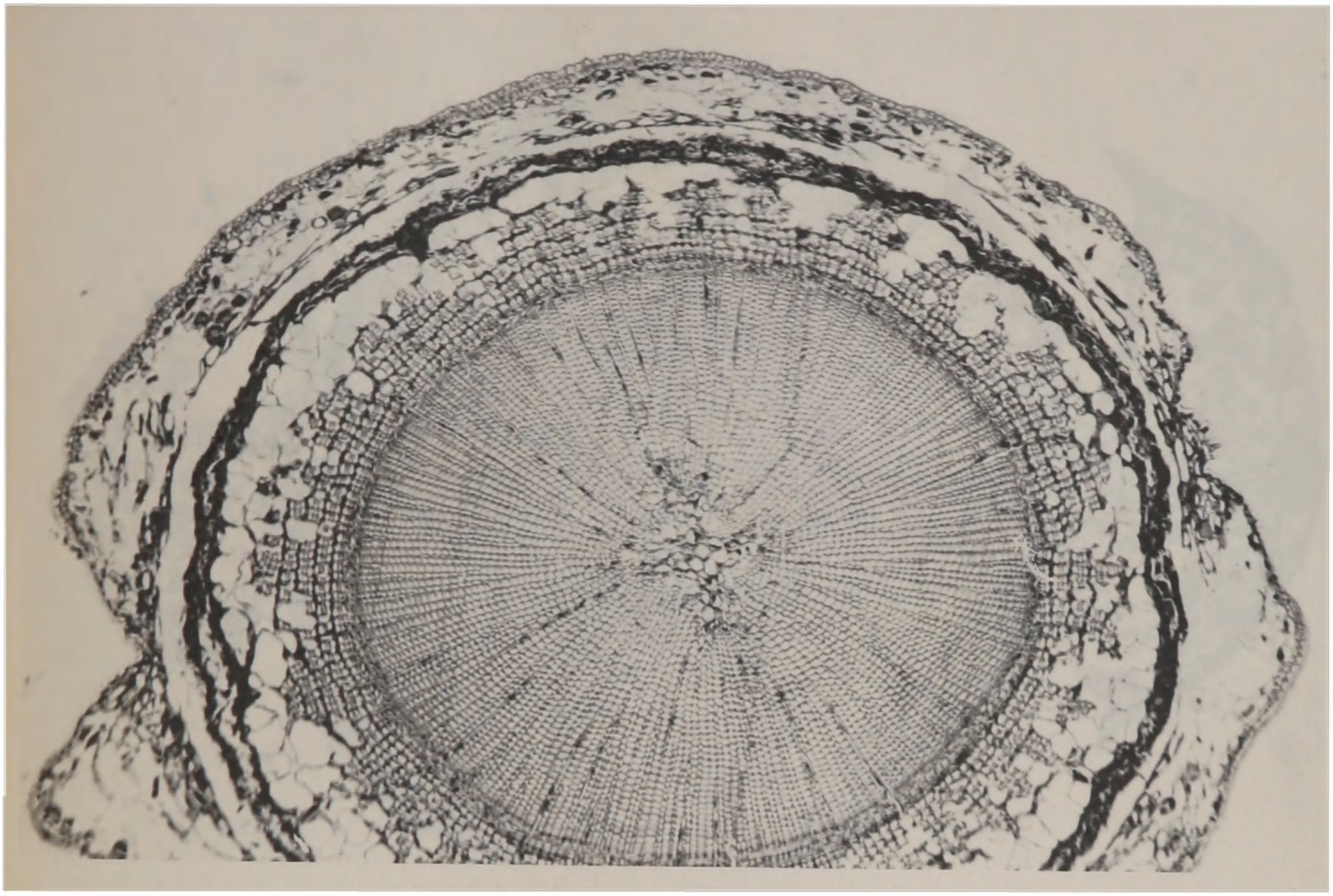

Figure 13. Transverse section of a mature stem and leaves of Thuja occidentalis cv. nigra ten cms from apex showing serious dehydration of tissue at 150°F (X80).

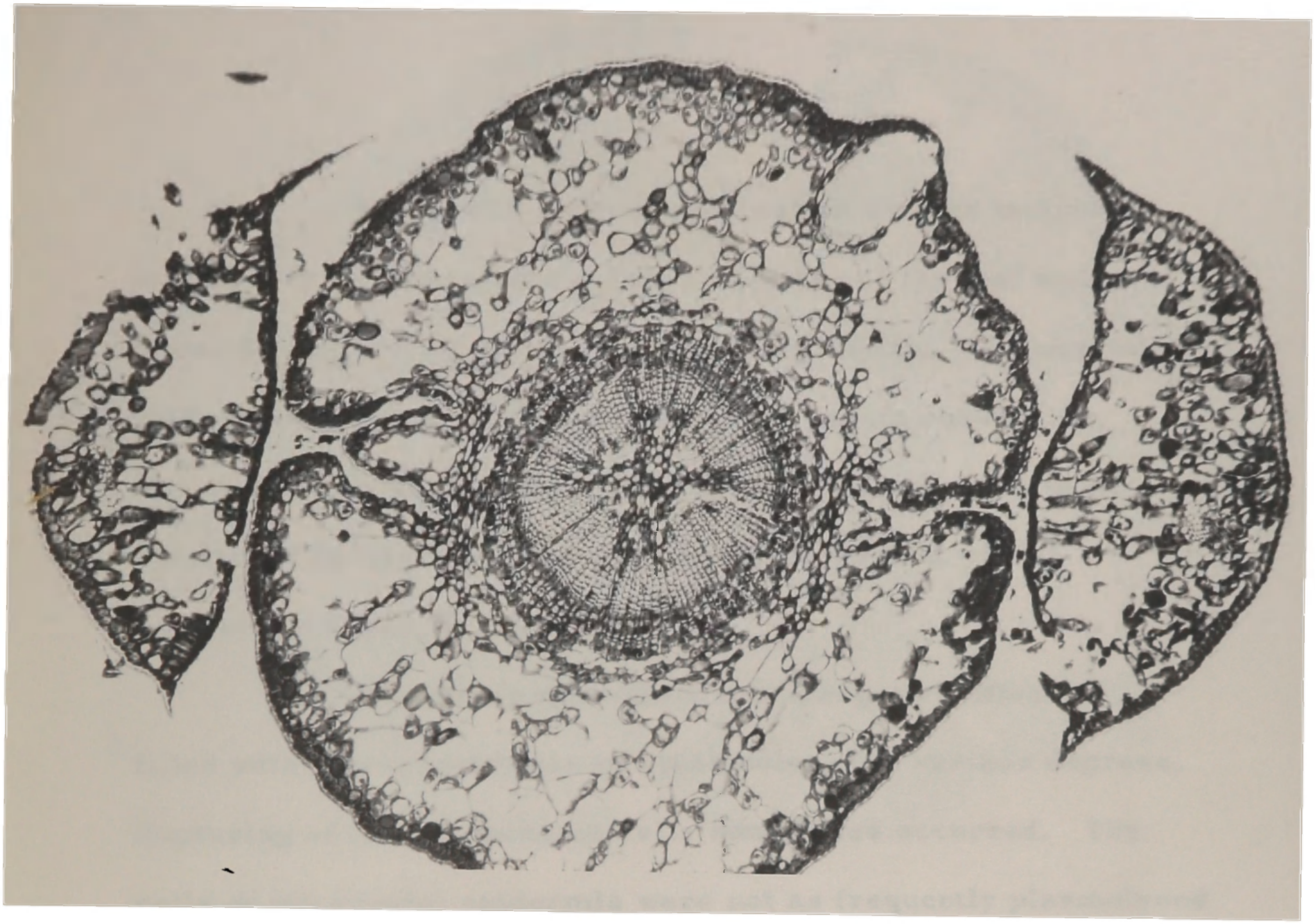

Figure 14. Transverse section of stem and leaves of Thuja occidentalis cv. nigra seven cms from the apex showing injury from freezing at 32°F (X80).

Artifically induced desiccation by high temperatures severely altered the cellular anatomy of the leaf and stem of Thuja occidentalis cv. nigra (Figure 15). The abaxial epidermal cells became depositories for tannin substances. The cells were atrophied, shrunken, and abaxial walls were convex. The hypodermal cell lumen were packed with tannin-like deposits (Figure 16).

Misshapen mesophyll was irregular, often filled with tannin materials and plasmolyzed in various degrees. Rupturing of the parenchyma cells sometimes occurred. The cells of the adaxial epidermis were not as frequently plasmolyzed although there seemed to be some tangential shrinkage. Some cells appeared almost globose, with the convex surface toward the periphery.

Parenchyma cells of the phloem as well as the xylem rays were partly shrunken, and often depositories for the tannin substances.

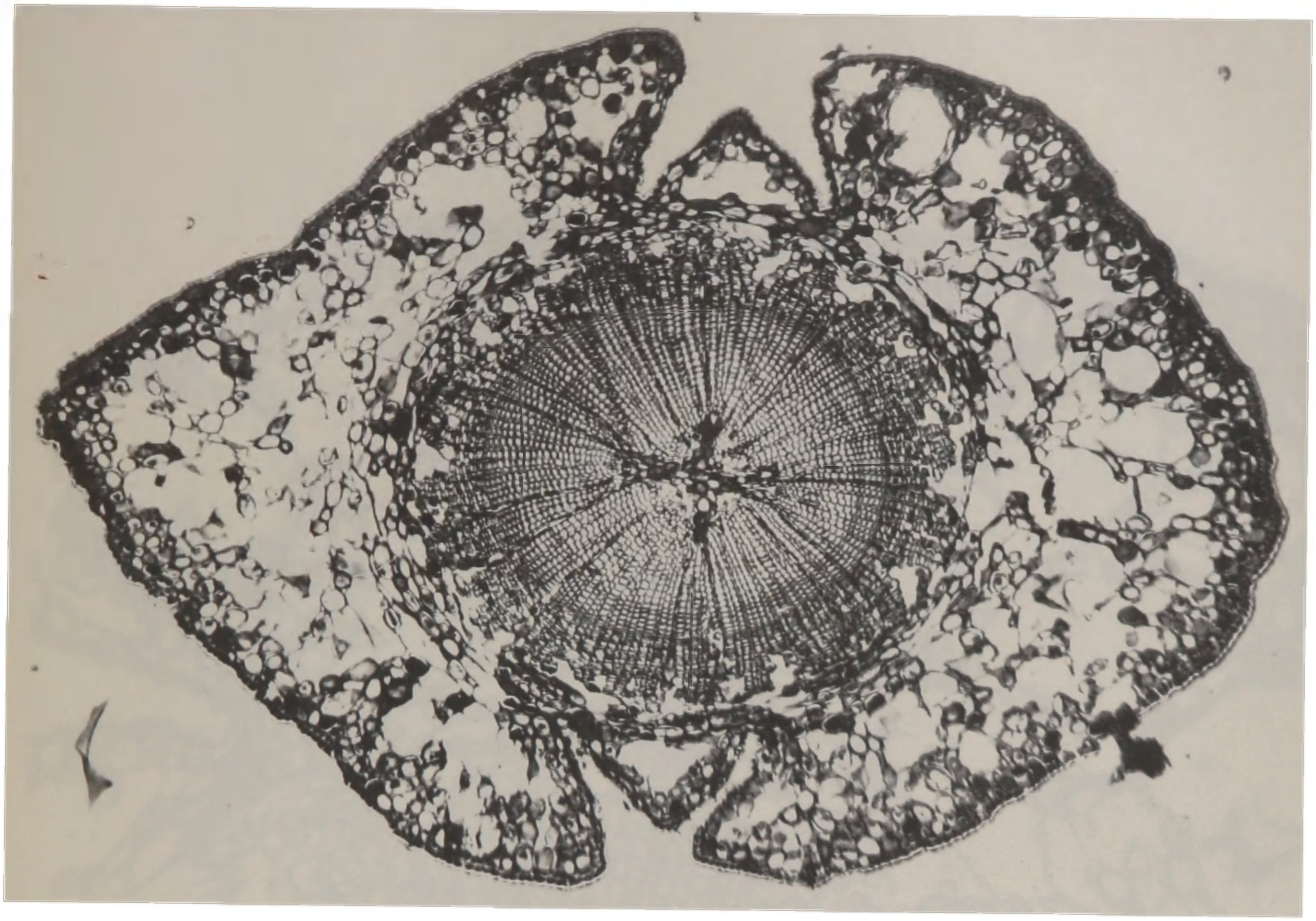

Figure 15. Transverse section of leaves and stem of Thuja occidentalis cv. nigra 10 cms from the apex of the stem showing injury from dehydration at 140°F in an oven (X80).

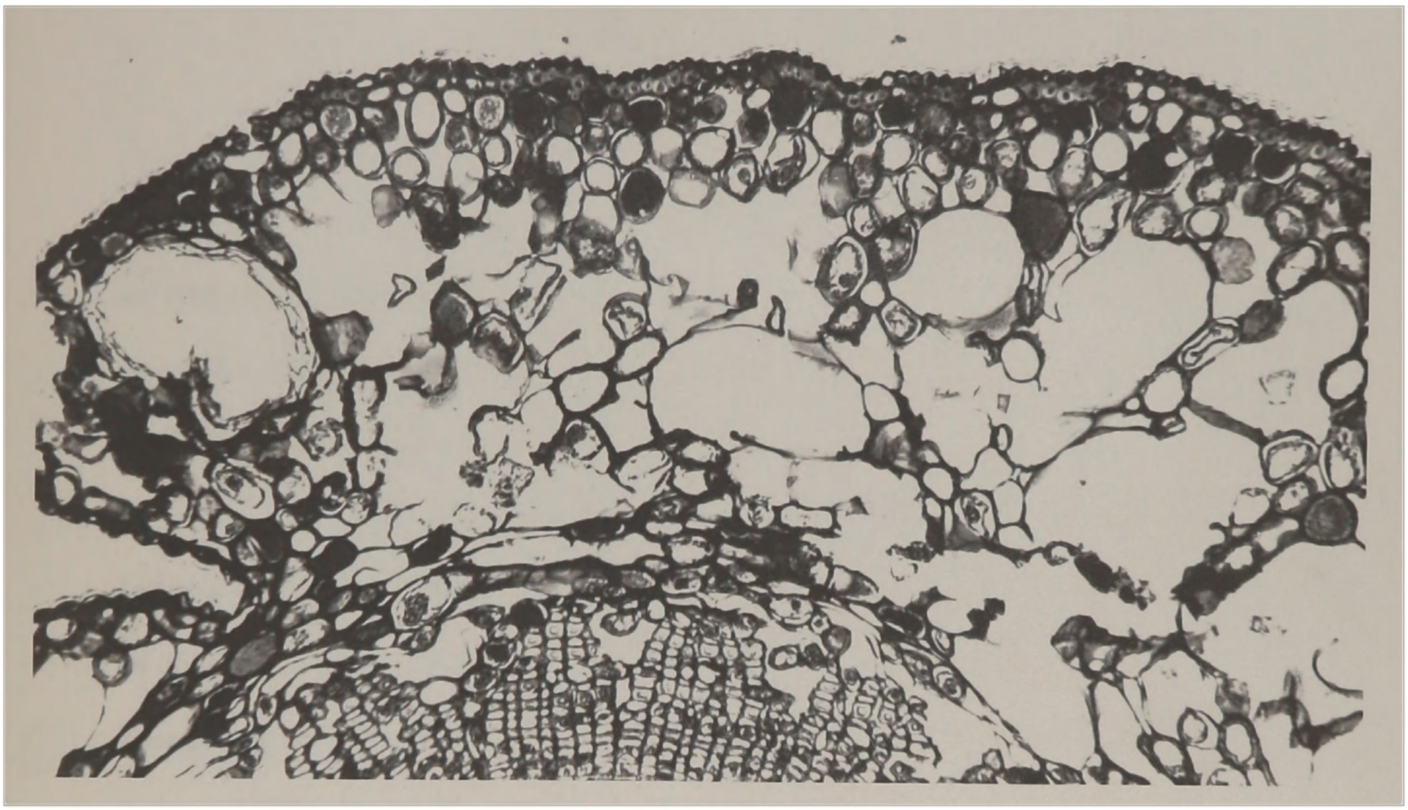

Figure 16. Transverse section of leaves and stem of Thuja occidentalis cv. nigra 10 cms from apex of the stem showing injury from dehydration at 140 ^{o}F in an oven (X240).

DISCUSSION OF RESULTS

From the results of the experiments attempting to improve growth following transplanting, it was not possible to make definite conclusions. No one method or one concentration was completely superior.

The vacuum method of application of growth regulators could be added to an already expanded list of techniques of application: Cholodny, 1936; Harrison, 1937; Block, 1938; Romberg and Smith, 1938; Stier and duBuy, 1938; Thimann and Lane, 1938; Howlett, 1939; Gustafson, 1940; Swartley, 1941; Zimmerman and Hitchcock, 1944; Withrow, 1945; Felber, 1947; Watson, 1948; Randhawa and Thompson, 1948; Mitchell, 1951; Doran, 1952; Nagato and Sato, 1952; and Asen and Hamner, 1953.

In general, total linear growth was retarded by the use of one ppm indoleacetic acid solution. Perhaps the growth regulator retarded the terminal portion of the stem, and consequently stimulated the growth of axillary buds. Support of this reasoning was offered by Zimmerman and Hitchcock (1940, 1942) using hormone treated *Hibiscus syriacus*;and *Rosa hybrida* cv. Better Times, Asen and Hamner (1953).

TABLE 9

Precipitation May to November, 1953 Compared to the 30-Year Average*

Month	Precipitation (Inches)	
	Average	1953
May	3.75	1.75
June	3.37	2.87
July	2.28	1.03
August	2.68	3.80
September	3.05	1.40
October	2.45	1.43

*From U. S. Weather Bureau local climatological data.

In these experiments and as pointed out by Gossard in 1943, there were, contrary to the findings of Swartley (1941) and Gregory, Imle and Camacho (1953), very few differences in the per cent survival of the transplanted plants.

The low concentration of indoleacetic acid may have stimulated the meristematic regions, particularly in the apical buds resulting in rapid and prolonged activity. In 1925, Snow demonstrated that *Phaseolus vulgaris* exhibited a positive correlation between the amount of growth substances applied and the activity of the cambia.

Since the plants received no supplementary irrigation during the period from May to November, the growth may have been reduced by a deficiency of water, as there was less precipitation in 1953 than the thirty year average (Table 9).

In addition to the different amounts of linear growth obtained for woody plants, as a result of treatment prior to transplanting, yields of *Lycopersicum esculentum* differed in number and weight of fruit (Tables 4, 5). In contrast to the findings of Stier and duBuy (1938), no significant differences were shown for the cultivar Golden Queen either in number or weight of early or total production. Results of Zimmerman *et al* (1944) and Withrow (1945), however, were in complete agreement with the results reported at this time.

The cultivar Red Cherry exhibited no significant differences in yield between the untreated and indoleacetic acid treated plants. Water treated plants produced significantly less fruit than the indoleacetic acid, but not the untreated plants. In weight, the water treated plants produced less fruit than either the untreated or indoleacetic acid treated plants.

Annual flowering plants exhibited a variety of responses (Tables 7 and 8). Immediately following treatment, stems bent and this persisted for a short period of time. Mitchell (1938) had observed this phenomena in _Phaseolus_, _Tagetes_, and _Coleus_. A delay of flowering of _Salvia splendens_ occurred as a result of the indoleacetic acid inhibition of the terminal growth, stimulating the axillary branches which were prevented from differentiation of floral primordia. Loss of the smaller buds as a result of the use of growth regulators was reported by Zimmerman and Hitchcock (1944).

The dehydration of leaves on unprotected plants has been attributed to high, rather than low temperature. The distinguishing characteristics of the injuries resulting from a range of temperature were that (a) at high temperatures, even for short periods, desiccation of the tissues resulted in a shrinkage of the cells of the epidermis, mesophyll and transfusion tissues; and (b) at low temperatures, rupturing and separating of the cells occurred, particularly those of parenchyma.

The rate of water loss from a plant has been calculated commonly by the determination of vapor pressure deficits as was done by Oosting in 1950. Vapor pressure deficit can be explained as the difference between the amount of water vapor actually present, and the amount that could exist without condensation when the temperatures of the air and evaporating surface were equal (Thornthwaite, 1940). Since this might be considered to be a direct indication of atmospheric moisture, independent of the temperature, it was believed to be much more indicative of the potential rate of evaporation. By using tables of vapor pressures (Table 10), it was possible to transform relative humidities to vapor pressure deficits from the known temperature. By erecting a cheesecloth enclosure, Batson (1933) was able to increase the relative humidity surrounding the plant by the cooling effect of shade and resulting reduction in rate and amount of transpiration.

From Figures 1, 2 and 3, the inverse relationship of temperature and relative humidity is apparent. The time of intersection was 1 to 2 hours earlier when recordings were obtained either under non-perforated or perforated polyethylene film. This was believed to have been caused by the solar radiation and the retarded movement of the air enclosed with the plant. From Table 10 using the temperatures and relative humidities, the vapor pressure deficit was determined. For the air the maximum vapor pressure deficit was 9.5, and for the perforated polyethylene approximately 33.7 mm of mercury. This deficit being greater, caused a movement of water from the plant. The rate of diffusion through the stomata was greater at higher temperatures.

Tables 11, 12, 13, and 14 show the temperatures of (a) open air, (b) soil--2 1/2 inch depth, (c) air under non-perforated, and (d) air under perforated polyethylene covers. The soil temperature fluctuated to a lesser degree than

Table 10. Vapor Pressure Deficit as Related to Relative Humidities at Different Temperatures*

Temperature		Vapor Pressure Deficits (Mm of Hg) at Given Relative Humidities										
		Relative Humidities										
C°	F°	100%	90%	80%	70%	60%	50%	40%	30%	20%	10%	0%
50	122	0	9.25	18.50	27.75	37.00	46.26	55.51	64.76	74.01	83.26	92.51
45	113	0	7.19	14.38	21.56	28.75	35.94	43.13	50.32	57.50	64.69	71.88
40	104	0	5.53	11.06	16.60	22.13	27.66	33.19	38.72	44.25	49.79	55.32
35	95	0	4.22	8.44	12.65	16.87	21.09	25.31	29.53	33.74	37.96	42.18
30	86	0	3.18	6.36	9.55	12.73	15.91	19.09	22.27	25.46	28.64	31.82
25	77	0	2.38	4.75	7.13	9.50	11.88	14.26	16.63	19.01	21.38	23.76
20	68	0	1.75	3.51	5.26	7.02	8.77	10.52	12.28	14.03	15.79	17.54
15	59	0	1.28	2.56	3.84	5.12	6.40	7.67	8.95	10.23	11.51	12.79
10	50	0	0.92	1.84	2.76	3.68	4.60	5.53	6.45	7.37	8.29	9.21
5	41	0	0.65	1.31	1.96	2.62	3.27	3.92	4.58	5.23	5.89	6.54
0	32	0	0.46	0.92	1.37	1.83	2.29	2.75	3.21	3.66	4.12	4.58

*From Oosting, 1950, p. 80

TABLE 11

Open Air Temperatures At Cottage Gardens Nursery, East Lansing, Michigan for 25 Days of March 1954.

Day	High	Low	Mean
1	29	22	25.5
	36	18	27.0
	34	17	25.5
	40	25	32.5
5	34	23	28.5
	41	19	30.0
	26	17	21.5
	40	20	30.0
	29	12	20.5
10	32	10	21.0
	39	15	27.0
	42	18	30.0
	49	18	33.5
	41	30	35.5
15	28	17	22.5
	31	13	22.0
	35	11	23.0
	47	23	35.0
	43	18	30.5
20	65	32	48.5
	33	17	25.0
	42	16	29.0
	48	19	33.5
	17	0	8.5
25	23	0	11.5

TABLE 12

Soil Temperature at 2 1/2 Inch Depth for 25 Days of March 1954

Day	High	Low	Mean
1	29	25	27.0
	30	24	27.0
	35	25	30.0
	35	27	31.0
5	42	27	34.5
	51	27	39.0
	28	25	26.5
	30	26	28.0
10	27	24	25.5
	34	22	28.0
	40	23	31.5
	44	25	34.5
	49	25	37.0
	37	27	32.0
15	32	26	29.0
	29	21	25.0
	30	23	26.5
	47	25	36.0
	40	25	32.5
20	55	31	43.0
	36	22	29.0
	46	23	29.5
	50	20	35.0
25	28	24	26.0

TABLE 13

Air Temperature Under Non-perforated Transparent Polyethylene Cover for 25 Days of March 1954

Day	High	Low	Mean
1	125	17	71.0
	124	11	67.5
	119	10	64.5
	119	14	66.5
5	116	12	64.0
	112	11	61.5
	45	10	27.5
	94	20	57.0
	94	13	53.5
10	116	2	59.0
	115	7	61.0
	103	8	55.5
	119	12	65.5
	48	27	37.5
15	63	15	39.0
	112	5	58.5
	100	3	51.5
	75	20	47.5
	82	14	48.0
20	76	32	54.0
	58	8	33.0
	96	10	53.0
	80	19	49.5
	35	0	17.5
25	137	0	68.5

TABLE 14

Air Temperature Under Perforated Transparent Polyethylene Cover for 25 Days of March 1954

Day	High	Low	Mean
1	105	15	60.0
	116	13	64.5
	106	11	58.5
	103	15	59.0
5	105	14	59.5
	105	10	57.5
	51	12	31.5
	80	20	50.0
	83	13	48.0
10	102	3	52.5
	98	9	53.5
	87	9	48.0
	96	15	55.5
	47	27	37.0
15	51	10	30.5
	93	4	48.5
	75	3	39.0
	64	19	41.5
	72	12	42.0
20	71	31	51.0
	34	17	25.5
	59	16	37.5
	50	19	34.5
	17	0	8.5
25	41	0	20.5

atmospheric temperatures. The mean temperatures under the polyethylene covers were consistently much warmer than in the open air. Temperatures under the transparent polyethylene covers of over $100^{o}F$ were not uncommon on bright clear days.

The soil temperatures during the period of injury were rarely much above freezing. Kramer (1940) concluded that the physiological activities of roots were greatly reduced by decreasing temperatures, especially the absorption of water. From the data of Roeser (1932), it was found that certain evergreens lost considerably more water by transpiration at a temperature of $51^{o}C$ than at $16^{o}C$.

The relatively low temperatures of the soil probably prevented the absorption of water by the plant in sufficient quantities and rapidly enough to replace that lost by transpiration.

It is believed that low temperatures of the soil, high air temperatures, and high vapor pressure deficits accounted for the desiccation injury to the plant. This was, in general agreement with the findings of Brierley and Landon (1946) and Maximov (1929).

Microscopic variations in the type of injury presented showed shrinkage and depositions resulting from high temperatures, a finding which is substantiated by the work of Simonds (1942) who reported marked splitting of the abscission layers in *Rubus* canes. Mix's work of 1916 found the youngest xylem cells were the first to be injured, contrary to what was observed in *Thuja*.

Lethal temperatures of living cells have been ascertained to range from 113 to 131^{o}F for most plants (Clum, 1926a). Higher temperatures were common under polyethylene covers during the experiment.

Increased metabolic activity of the plant caused by the higher temperature may have been a cause of injury. While the period of time a plant was capable of withstanding injury brought about by high respiration rates, high air temperatures and high transpiration rates was not determined, it seemed likely that a period of 1 to 2 days would be a maximum.

The use of transparent polyethylene film as a protective cover for ornamental plants cannot be recommended to replace the screens suggested by Thompson (1940). Although transparent polyethylene was found by Sweet (1953) to be an effective means of conserving moisture in potted plants, it was not satisfactory for outdoor trees and shrubs because extremely high temperatures developed beneath.

Transparent polyethylene film in the field deteriorated to the extent of splitting when exposed to climatic conditions of Michigan during February and March of 1954.

SUMMARY

Different transplanting techniques were studied to evaluate the effects of additional water and one ppm indole-acetic acid solution, taken up by the plant as a result of evacuation of air or soaking the roots. Using the amount of linear growth and development of lateral branches of *Syringa vulgaris* cv. Thunberg, *Forsythia spectabilis*, and the yield of fruit of cultivars Golden Queen and Red Cherry of *Lycopersicum esculentum* contrasts in growth were recorded.

Transparent polyethylene film covers (non-perforated and perforated by two 6-mm openings per square foot) were used in an effort to prevent injury of *Thuja occidentalis* cv. nigra under field conditions. Temperature fluctuations were determined from thermocouples attached to a "Minneapolis-Honeywell Brown electronik" continuous temperature recorder, and relative humidities by "Bendix-Friez" hygrothermographs.

Stem samples were collected from randomized locations on untreated, non-perforated and perforated transparent polyethylene covered plants in the field, killed in formalin-acetic-alcohol, dehydrated with tertiary-butyl alcohol, embedded in paraffin, sectioned eight micra in thickness, and stained with

safranin and anilin blue. Sections from apparently uninjured branches, one-half stored dry, the remainder in tap water were subjected to varying lengths of time (15, 30, 60 and 120 minutes) and temperatures 32 to 140°F before the preparation of slides.

Growth changes were not uniform or consistent for all plants. The number of branches on treated plants of *Syringa* was increased, whereas *Forsythia* was decreased by treatment. *Lycopersicum esculentum* cv. Golden Queen showed no significant differences in number or weight of either early or total production. The cultivar Red Cherry, however, when treated with water produced significantly fewer fruit than either the untreated or indoleacetic acid treated plants.

Likewise, certain annual flowering plants exhibited a variety response. Flowering of *Salvia splendens* was delayed by indoleacetic acid treatment.

Temperatures of the air and temperatures under polyethylene film were found to fluctuate widely during March 1954. Temperatures of unprotected plants were consistently lower than those of the covered plants. A reduction of temperature of from 1 to 3°F was observed under perforated as contrasted with non-perforated polyethylene film. Soil temperatures remained near freezing during the month of March.

Injury of two types occurred: (a) desiccation accompanied by browning; and (b) desiccation without browning, usually being light green or yellow and brittle. Relative humidities fluctuated from 100 per cent during the periods of lowest temperatures to approximately 20 per cent during periods of higher temperature.

Leaves of Thuja occidentalis cv. nigra possessed a heavy cuticle. The epidermis was a single layer of cells with rather heavy walls. The peripheral walls were generally convex. Fewer stomata were found on the abaxial surface than on the adaxial surface. The hypodermis, composed of collenchyma cells, was adjacent to the epidermis. The mesophyll was composed of irregularly shaped parenchyma cells filled with chloroplasts. There were large intercellular spaces. The vein composed of adaxial xylem and abaxial phloem was surrounded by transfusion tissue, which in turn was bounded by a single-layered heavy-walled endodermis.

Stomata were sunken, bounded by subsidiary cells which arched over the somewhat lignified guard cells. In older leaves, one or more resin ducts were located on the abaxial side of the leaf. In older stems, pith parenchyma possessed heavy cellulosic walls. Ray cells were in uniseriate rows, elongated toward the periphery of the stem.

Injury on uncovered plants originated on south or southwest exposures of the plants. The epidermal cells are inactivated by deposition products. Palisade-like cells and mesophyll cells appeared shrunken, and possessed large intercellular spaces. Xylem cells were little affected, but the transfusion tissues were collapsed. Endodermal cells were slightly shrunken.

Under the transparent polyethylene covers, the leaves were first pale green, yellow and finally brown. Cellular contents of all tissues were atrophied, with dense tannin materials being deposited in the epidermal and hypodermal cell lumens. All parenchyma cells, regardless of location in the leaf or stem were shrunken, contained deposits of tannins and were misshapen.

The cells of the cuticle, epidermis and hypodermis of leaves and stems, which had been artifically frozen, were not observed to differ from uninjured tissues. The location of parenchyma cells was altered, some plasmolysis occurred, but principally injury was observed as rupturing and separating of cell walls.

Artifically injured leaves and stems possessed atrophied cells in the epidermis, endodermis, and parenchyma cells regardless of location. Depositions of tannin materials were prevalent in all of these cells.

Observations showed that injuries, induced artifically by freezing, were not of the same type or magnitude as those produced artifically by high temperature desiccation. Transparent polyethylene film was an unsatisfactory cover for woody plants because of high air temperature developing within the cover, and because of the rapid disintegration of the plastic. Injuries of untreated plants, plants covered with transparent perforated and non-perforated polyethylene film were similar to those produced artifically by high temperature.

Sixteen figures and fourteen tables were included.

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