

REVIEW OF TECHNIQUES USED TO EVALUATE

SEEDLING QUALITY ¹

Annabelle Jaramillo²

ABSTRACT

The increasing need for high quality conifer planting stock has increased interest among nursery managers, silviculturists and researchers. Measurement of electrical, chemical and other characteristics that have been used to evaluate planting stock are reviewed.

INTRODUCTION

The increasing need for conifer planting stock of high quality has increased interest among nursery managers, silviculturists, and researchers in improving methods of evaluating the quality of planting stock. Nursery managers want to determine when planting stock is ready to be lifted, stored, or shipped to the planting location. They want to know how well the stock is doing in its new growing environment and to be able to relate its success or failure to cultural or handling practices at the nursery. Rising costs of equipment, chemicals, and payrolls increase a manager's need to use human and fiscal resources more efficiently. Users (silviculturists, reforestation specialists, and others) want to make knowledgeable decisions about when, where, and how to use the planting stock. They need to know the importance of timing cultural practices and of site-matching. They must know whether stock of questionable quality should be used or discarded. Furthermore, nursery managers and users need to understand each other to assure successful reforestation, and knowledge of stock quality is an important part of this communication. Such knowledge and understanding of nursery and field practices can increase the ability of both the nursery manager and user to make responsible decisions.

Researchers have directed much interest toward gathering biological data that can help the nursery manager and user in evaluating stock quality. Sutton (1980) has reviewed some of this research presented at a workshop focusing on evaluation of planting stock quality, held as part of an International Union of Forestry Research Organizations (IUFRO) workshop in New Zealand in 1979.

What is stock quality? Substantial confusion exists on this question among nursery managers, users, and researchers. The term is often used loosely: some use it to describe whether a seedling is dead or alive; others mean whether seedlings are ready to handle (i.e., whether they are in the "right" condition); still others use the term to describe whether or not seedlings are physiologically and morphologically adapted to grow on specific sites. Thus, to improve communication and subsequently

¹ Paper presented at the joint meeting of the Western Forest Nursery Council and Intermountain Nurseryman's Association, Boise, Idaho, August 12-14, 1980.

²Botanist - Pacific Northwest Forest and Range Experiment Station USDA Forest Service Corvallis, Oregon.

increase reforestation success, nursery managers, users, and researchers must clearly define "stock quality"--or at least identify what particular aspect applies for a given use.

Efforts to evaluate planting stock quality have taken many directions. Seedling physiology has been evaluated by physical parameters, such as measurements of plant water status (Cheung et al. 1975; Cleary 1971; Cleary and Zaerr 1979), electrical impedance (Glerum 1970, 1973, 1979; van den Driessche 1969, 1973a, 1976), resistance (Ferguson et al. 1975), and conductivity (Aronsson and Eliasson 1970). Quantitative measures of carbohydrate reserves (Krueger and Trappe 1967) and mineral-nutrient content (Krueger 1967; van den Driessche 1971, 1973a, 1980) have been used to describe chemical parameters. Other tests of seedling quality have included measurements of field survival and laboratory tests of survival potential (Jenkinson and Nelson 1978; Askren and Hermann 1979; Hermann and Lavender 1979).

In this paper, I will review several methods currently being used to evaluate quality of conifer planting stock. I am omitting evaluation of stock quality by morphological characteristics and the matching of stock to specific planting sites because of time limitations. Instead, I will concentrate on measures of seedling health and vigor.

DESCRIPTIONS

Tests or techniques for estimating seedling quality can be used at different levels of refinement; some may simply tell us whether seedlings are dead or alive. In areas subject to heavy frosts, estimating cold hardiness in seedlings is useful. Estimating levels of dormancy can assist the nursery manager in choosing the best lifting times. Some tests take weeks to provide information, but others can give immediate answers. No one test can tell us all we may wish to know about the seedling. We must be cautious when we use instruments as predictors of seedling quality. For example, a seedling can be dead and still have low moisture stress. We should know about differences in values that occur at different times and how to interpret these data.

ELECTRICAL MEASUREMENT OF PLANT TISSUES

Cell-wall resistance, cytoplasm resistance, and cell-membrane resistance and capacitance are components of electrical circuits in plants. Changes in the physiological status of a plant can effect changes in these components. Because electrical resistance in plant tissues is ionic rather than electronic, measures of these characteristics in plants must be interpreted carefully; variation can be influenced by factors such as seedling diameter, temperature, and moisture. The following techniques have been suggested as tools that nursery managers can use to evaluate the physiological status of seedlings.

Electrical impedance. Electrical impedance is measured by passing electrical current through a four-electrode probe inserted into plant tissue, usually at two frequencies, and expressed as a ratio. Measurements of electrical impedance have been used in studies of frost hardiness (Aronsson and Eliasson 1970; Glerum 1973; van den Driessche 1969, 1973b, 1976). In these studies, impedance measurements were made before and after plant tissues were subjected to freezing temperatures. Frost hardiness was determined by the extent of injury to the tissues, which correlated with a significant decrease in electrical impedance. Although electrical impedance measurements are nondestructive and easy to make, they are not easily translated into physiological condition of plant tissue. Electrical impedance measurements are affected by stem diameter, temperature, and tissue moisture content.

Glerum (1979) indicated that impedance measurements are of limited use in studies of water potential. Kitching (1966) also found electrical impedance measurements were not useful as an index of moisture stress. Measurements of electrical impedance appear to be promising in some areas of study, but because they vary by species and location they will be useful only when extensively studied and calibrated locally (i.e., in each nursery) for each species grown. A IUFRO project group focusing on nursery problems is currently suggesting standardization of electrical impedance curves for each nursery.

Oscilloscope square-wave apparatus. The application of a square-wave electrical pulse without seedling stem tissue and observation of trace forms on an oscilloscope (fig. 1) has been suggested as a tool to evaluate the condition of plant tissue



Figure 1. Oscilloscope/square-wave apparatus. Insert: electrode from oscilloscope/square-wave apparatus in seedling terminal.

(Zaerr 1972; Ferguson et al. 1975). Zaerr exposed plant tissues to freezing, steaming, and herbicides. He found that trace forms of healthy tissues differed from those of dead tissues. Although the species being observed had their own characteristic trace forms, "...differences in shape of curve for live and dead tissues of a given species were consistent." Ferguson and others (1975) noted changes in trace form (Fig. 2) of several species at different times of the year and suggested that the forms could be used to determine dormancy. These changes appeared to be related to the growth activity or dormancy of their samples. Dormancy was "measured" by time of year and not actually determined by growth tests, however. In a previous paper (Jaramillo 1978), I reported a lack of correlation between "dormant" trace forms and cold hardiness of Douglas-fir seedlings. I found that visual observation of changes in trace form could not be used as indicators of cold hardiness. Askren and Hermann (1979) took voltage measurements at three constant points of the trace--high-frequency

(HFV), mid-frequency voltage (MFV), and low-frequency voltage (LFV)--and used ratios of these measurements to typify trace forms. These ratios were used in tests of seedling survival potential. They found that "...trace character apparently does not indicate vigor as such, and thus is poorly suited for predicting survival potential." In her investigation of the relation of electrical impedance to vegetative maturity and dormancy in red-osier dogwood, Parmelee (1979) ruled out the oscilloscope/squarewave technique because of the difficulties encountered with interpretation and reliability of the square wave. Standard wave-traces would have to be established for each plant species because each tends to exhibit its own characteristic traces. In addition, slight movement of a twig in which the probes are inserted creates differences in wave form. These limitations cast doubt on a practical use of squarewave forms as visual indicators of plant physiological condition.

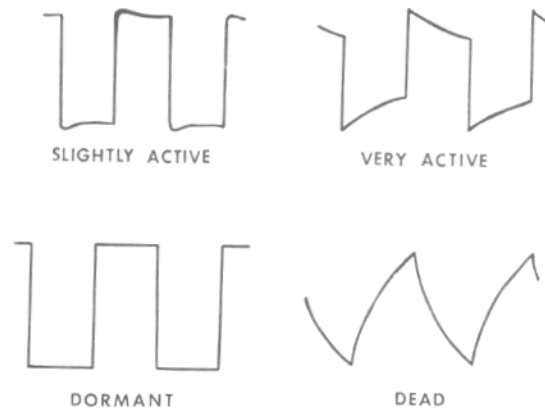


Figure 2. Square-wave trace forms indicating growth condition (Ferguson et al. 1975).

Dormancy meter. A solid-state dormancy meter (Fig. 3) has been designed for determining physiological activity of nursery stock, as a less costly substitute for the oscilloscope/square-wave apparatus. The Missoula Equipment Development Center's engineers have determined "...that an instrument which measures the ratio of voltage at 500 hertz and at 10 kilohertz gives basically the same performance..." as the oscilloscope/square-wave apparatus. Preliminary tests⁴ that I made comparing dormancy meter readings with square-wave trace form showed little or no correlation between the two instruments. I did not compare meter readings with actual seedling physiological status. A more thorough investigation of this relationship should be made before the instrument is recommended for operational use.

³February 1977. Report 7741 2505. Equipment Development Center, USDA-FS, Fort Missoula, Missoula, Montana.

⁴Jaramillo, Annabelle E. 1978, 1979. Office Reports: Comparison of Oscilloscope/Square Wave Apparatus and the MEDC Dormancy Meter.

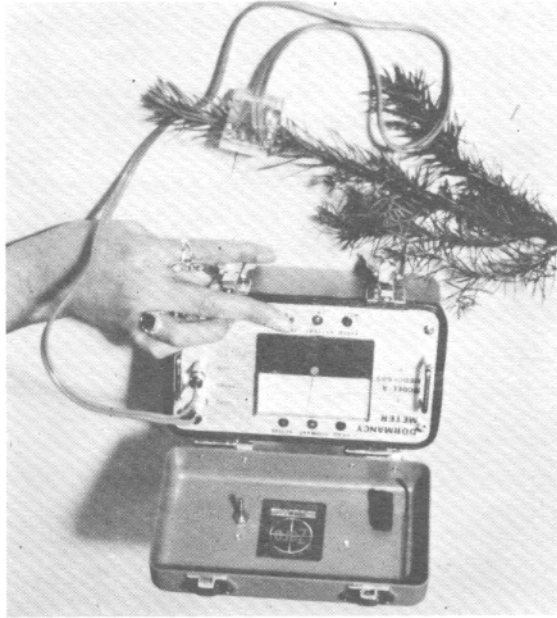


Figure 3. Dormancy meter designed by Missoula Equipment Development Center, USDA Forest Service.

PHYSIOLOGICAL WATER STATUS

Energy status of water in plants results from transpiration, evaporation, and other factors. Water status in a soil-plant continuum is dynamic, and the water is rarely in equilibrium with that in surrounding areas. Because the process is dynamic, measurements must be interpreted carefully by someone knowledgeable in plant-water relations.

Because plant responses to water stress are closely related to the energy required to remove a unit of water from soil, instruments used to measure soil-water status have been investigated as possible tools to estimate plant-water status. I will describe only techniques that measure water inside plants, however.

Psychrometric measure of water potential. Water potential of a small sample of plant tissue is measured by condensing water from the atmosphere in a psychrometer chamber, in which the tissue is placed, on to a measuring junction (thermocouple). Measurement of output voltage (on a microvoltmeter connected to a psychrometer) across the thermocouple is a function of the water potential in the psychrometer and ambient temperature. Stein and Jaramillo⁵ tested water potential of Douglas-fir needles and found that the psychrometric measurements were too variable to be used as indicators of seedling quality. The sensitivity of the psychrometer (fig. 4) to fluctuating ambient temperatures, the lengthy periods needed to calibrate the instrument and make observations, and the need to establish standard curves for the tissues being tested make it highly impractical as a predictive tool.

⁵Stein and Jaramillo, unpublished data, on file at FSL.



Figure 4. Equipment needed for water-potential studies. Psychrometer chamber apparatus is connected to a microvoltmeter. A paper disc, saturated in tissue water extracted by maceration is inserted in the psychrometer chamber. The equipment is calibrated by known standards.

Freezing-point depression. Carey and Fisher (1969) and Fisher (1972) have described a small, portable instrument that measures the freezing-point depression of plant tissue. It consists of a small freezing chamber mounted on the cold side of a Peltier battery. They suggest that "...freezing-point depression measured immediately after ice crystals begin to form in plant tissue in the field could give on-the-spot estimates of plant water stress." This has advantages over the psychrometric method because it is less time consuming, less expensive, and can be used on more types of plant tissue. The practicality of both water-potential and freezing-point depression measures need to be investigated more intensely for conifer seedling tissues before they can be suggested as useful tools for evaluating stock quality.

Pressure-chamber (pressure-bomb) technique. At a recent IUFRO Conference, Cleary and Zaerr (1979) described the pressure-chamber technique used to evaluate plant water status. In the instrument (fig. 5), the negative potential in the xylem of the plant is balanced with the positive pressure of a chamber. A sample is cut from a seedling and placed in the chamber with the cut end exposed through the chamber cover. Pressure is slowly increased in the chamber until water is forced back to the cut surface. This pressure is an estimate of plant moisture stress (PMS) in bars. Drawbacks of the technique are that values of zero stress levels are not obtainable and that it requires compressed gas, which can be potentially dangerous if not used carefully. The pressure chamber technique is relatively fast and easy and gives good estimates of seedling water status. But it should not be the only tool used to determine stock quality, because water status does not always tell the whole story. The pressure chamber is currently being used at many nurseries in the Pacific Northwest for regulating irrigation schedules and determining when stress is too high for lifting and handling. Many silviculturists also use pressure chambers for checking stock quality in the field.

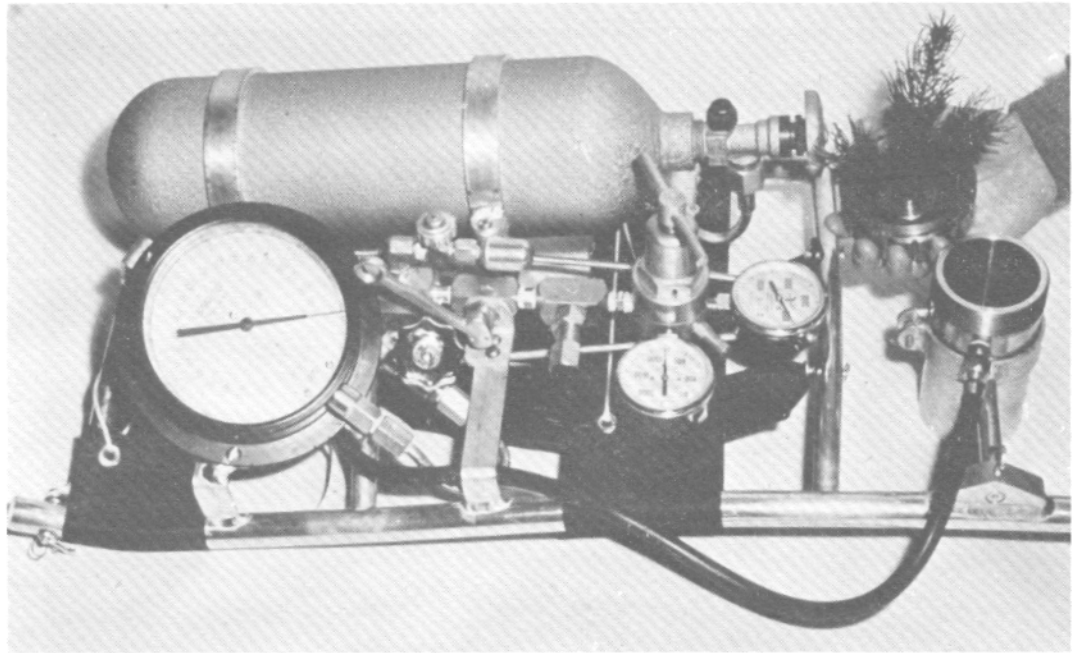


Figure 5. Pressure-chamber apparatus used to measure moisture stress using compressed gas.

Hydraulic Press. A hydraulic press has been developed (fig. 6) for measuring leaf-water stress and soil-water content. Measured hydraulic pressure beneath a flexible membrane is used to press a leaf or other plant tissue against a plexiglass window. As pressure is applied, water appears at the cut edge of the stem or leaf. Additional pressure causes changes in leaf color. Water stress is characterized by the pressure required to produce the color change accompanied by continual water excretion. Advantages are that the instrument can make measurements rapidly and that it does not require a compressed gas supply. Disadvantages reported by Cleary and Zaerr (1979) are: the end point is indefinite; precision is low; the instrument must be calibrated with a pressure chamber; foliage of different ages gives different results; and a very large sample size is required for acceptable accuracy. Until further data on the hydraulic press are reported, operational use of the technique is questionable.



Figure 6. Hydraulic press used to measure moisture stress.

CHEMICAL CHARACTERISTICS OF PLANT TISSUES.

Foliage nutrient content and carbohydrate reserves have been the most commonly studied chemical characteristics of seedlings. Visual clues--such as needle chlorosis, needle curl, and stunted growth--can tell us that some mineral element vital for growth is deficient. Knowledge of the mineral nutrient status of seedlings can tell us whether to apply or delete fertilizers to insure optimum growth of seedlings. Similarly, knowing something about carbohydrate levels at different times of the year helps establish handling practices that take advantage of carbohydrate reserves within the seedlings. We must know how to interpret the data and how they relate to planting stock quality, however.

Foliage nutrient content. Foliage nutrient content has been suggested as a predictor of seedling survival. Wakely (1949) suggested that evidence of a direct relation "...between chemical (nutrient) differences in seedlings..." and "...differences in survival and growth after planting..." was needed to establish foliage nutrient content as a predictor. Switzer and Nelson (1963) found that for 1+0 loblolly pine seedlings, a linear relation existed between height at 3 years in the field and foliar nitrogen content at lifting. Regression analyses indicated, however, that field survival could not be predicted by foliage nutrient content. Ileiner and Lavender (1972) found that foliar calcium/potassium ratios of 2+0 Douglas-fir seedlings did not correlate with field survival. In their investigation of 1+0 seedlings of several pine species, Gilmore and Kahler (1965) found no relation between field survival and foliage contents of nitrogen, phosphorus, or potassium. Although nursery fertilization does enhance seedling growth, foliage nutrient content at time of lifting has not been useful as a predictor of field survival. In addition to a lack of correlative data, assessment of foliage nutrient content is further hampered by the need for specialized equipment, a skilled investigator, and lengthy lapse times for results. Although many studies have been aimed at determining desirable nutrient levels within seedlings, no standardization of these levels and few correlations with field performance have been made. These factors limit the use of nutrient status as a predictor of stock quality.

Carbohydrate reserves. Krueger and Trappe (1967) observed substantial seasonal changes in carbohydrate concentrations of Douglas-fir seedlings at the USDA Forest Service Wind River Nursery. In the fall when top growth stopped, sugar reserves gradually increased. Maximum concentrations occurred with the coldest weather. A late winter decrease in sugar concentrations coincided with an increase in starch concentrations. They suggested that information on carbohydrate reserves might be used in selecting appropriate times to handle planting stock, which might improve survival in the field. Like mineral nutrient content, carbohydrate reserve data are useful only if we have standards for comparison. Further work is needed before information on carbohydrate reserves in conifer seedlings can be used as a predictive tool. The need for specialized equipment, skilled personnel, and lapse times for results also makes operational use doubtful.

GROWTH-EVALUATION TECHNIQUES

Jenkinson and Nelson (1978) reported the use of root-growth capacity (RGC) of Douglas-fir seedlings as a predictor of field survival. RGC, previously termed root-regenerating potential (RRP), is the seedling's ability to initiate new roots and elongate existing roots under conditions favorable for growth. RGC is estimated by measuring new root growth in controlled favorable environments for a specific period. Jenkinson and Nelson (1978) found that field survival was associated with RGC values. RGC appears promising as one method of evaluating quality of planting stock. Additional standards of RGC for different species, seed sources, and nurseries must be established and correlated with field performance, however.

Hermann and Lavender (1979) encouraged testing of seedling vigor as a measure of stock quality. This requires maintaining seedlings under constant conditions in a growth room and observing bud flush and survival. A random sample is selected from a nursery lot for testing. Half of the sample is kept as a control (not stressed), the other half is stressed by exposing bare roots and shoots to 90°F and relative humidity of 30 percent for 15 minutes just before potting the seedlings. (This stress testing can be varied according to the intended planting site.) All seedlings are potted and then kept for at least 4 weeks at 70°F (t5°F) with a 16-hour photoperiod of 500 foot candles. In 4-6 weeks, bud flush can be used as an indicator of vigor. An additional sample should be monitored in the field. Hermann and Lavender suggest that if a lot is deemed satisfactory in vigor tests but has poor survival in the field, seedlings may not have been properly handled during storage, transport, or planting. If field survival is much better than in the growth room, the procedures for that particular vigor test may have been conducted improperly. Poor survival in the field and the growth room can indicate a problem in the nursery environment or cultural practices.

DISCUSSION

I believe the most successful evaluations of planting stock now available are tests of seedling vigor and field survival. Although they do require time, they integrate all the various conditions and quality factors and give us information that can be useful for suggesting future nursery and planting practices. We can include other types of testing to determine specific characteristics and to search for more rapid determiners of overall seedling quality.

Coniferous tree seedlings--like all plants--are complex systems in which many internal processes occur and interact. These systems are affected by the environment in which the seedlings grow. A multitude of studies is directed at unraveling and understanding these complex systems. The techniques I have reviewed here are a small part of these investigations. Measures of the physical properties of seedlings have been attempted through studies of electrical impedance, resistance and capacitance of seedling cells and tissues, and plant moisture status. Analyses of foliage nutrient content and carbohydrate reserves have given us much information about the biochemical nature of seedlings. Tests of growth and seedling vigor are important for evaluating stock quality.

We should not depend on one method alone to tell us everything we want to know about seedlings. We have to look at what a combination of methods tell us and continue to look for new tools to evaluate planting stock quality. My fantasy is to have, some day, a tricorder (the little black box carried by Mr. Spock, science officer on the Starship Enterprise) to carry into a greenhouse or out to the nursery bed and have it tell me, at the push of a button, all there is to know about a seedling and its potential. In reality, we do not have such a single tool--and probably never will. But the tools we do have can be used separately or jointly to gather the information we need about planting stock to improve our reforestation efforts.

REFERENCES CITED

- Aronsson, A., and L. Eliasson.
1970. Frost hardiness in Scots pine (Pinus sylvestris L.) I. Conditions for test on hardy plant tissues and for evaluation of injuries by conductivity measurements. Stud. For. Suecica. 77, 30 p.

- Askren, Catherine, and Richard K. Hermann.
1979. Is the oscilloscope technique suitable for predicting survival of planting stock? *Tree Plant. Notes.* 30(4):7-11.
- Cary, J. W., and H. D. Fisher.
1969. Plant moisture stress: A portable freezing-point meter compared with the psychrometer. *Agron. J.* 61:302-305.
- Cheung, Y. N. S., M. T. Tyree, and J. Dainty.
1975. Water relations parameters on single leaves obtained in a pressure bomb and some ecological interpretations. *Can. J. Bot.* 53:1342-1346.
- Cleary, Brian D.
1971. The effect of plant moisture stress on the physiology and establishment of planted Douglas-fir and ponderosa pine seedlings. Ph.D. Thesis, *Oreg. State Univ., Corvallis.* 85 p.
- Cleary, Brian, and Joe Zaerr.
1979. Use of the pressure chamber technique to monitor and evaluate seedling water status. IUFRO workshop: Techniques for evaluating planting stock quality. August, 1979. Rotorua, Christchurch, New Zealand. 13 p.
- Ferguson, Robert B., Russell A. Ryker, and Edward D. Ballard.
1975. Portable oscilloscope technique for detecting dormancy in nursery stock. USDA-FS Gen. Tech. Rep. INT-26. 16 p.
- Fisher, H. D.
1972. An inexpensive method of determining plant moisture stress using freezing-point depression. *Soil Sci.* 113(6):383-385.
- Gilmore, A. R., and L. H. Kahler.
1965. Fertilizers and organic additives in pine nursery seed beds: Effect on field survival, field growth, and chemical content of foliage. *Tree Plant. Notes.* 73:21-27.
- Glerum, C.
1970. Vitality determinations of tree tissue with kilocycle and megacycle electrical impedance. *For. Chron.* 46(1):63-64.
- Glerum, C.
1973. Annual trends in frost hardiness and electrical impedance for seven different coniferous species. *Can. J. Plant Sci.* 53(4):881-889.
- Glerum, C.
1979. Electrical impedance techniques in physiological studies. IUFRO workshop: Techniques for evaluating planting stock quality. August, 1979. Rotorua, Christchurch, New Zealand. 15 p.
- Heiner, Terry D., and Denis P. Lavender.
1972. Early growth and drought avoidance in Douglas-fir seedlings. Res. Pap. 14, *For. Res. Lab., Oreg. State Univ., Corvallis.* 7 p.
- Hermann, Richard K., and Denis P. Lavender.
1979. Testing the vigor of coniferous planting stock. Res. Note 63, *For. Res. Lab., Oreg. State Univ., Corvallis.* 3 p.

Jaramillo, Annahelle.

1978. An integrated study of nursery stock conditioning: Oscilloscope readings and cold hardiness. Conf. and Workshop Proc., October 1978, West. For. Nursery Council. and Intermt. Nurseryman's Assoc., Eureka, Calif., 1978. p. B106-B116.

Jenkinson, James L., and James A. Nelson.

1978. Seed source lifting windows for Douglas-fir in the Humboldt Nursery. Conf. and Workshop Proc., October 1978, West. For. Nursery Council. and Intermt. Nurseryman's Assoc., Eureka, Calif., 1978. p. B77-B95.

Kitching, R.

1966. Investigating moisture stress in trees by an electrical resistance method. For. Sci. 12:193-197.

Krueger, Kenneth IV.

1967. Foliar mineral content of forest- and nursery-grown Douglas-fir seedlings. USDA For. Serv. Res. Pap. PNW-45. 12 p.

Krueger, Kenneth W., and James M. Trappe.

1967. Food reserves and seasonal growth of Douglas-fir seedlings. For. Sci. 13(2):192-202.

Parmelee, Kathleen.

1979. The relationship of electrical impedance to vegetative maturity and dormancy in red-osier dogwood. Masters thesis. Oreg. State Univ., Corvallis. 87 p.

Switzer, G. L., and L. E. Nelson.

1963. Effect of nursery fertility and density on seedling characteristics, yield, and field performance of loblolly pine (*Pinus taeda* L.). Proc. Soil Sci. Soc. Am. 27:461-464.

Sutton, R. F.

1980. Techniques for evaluating planting stock quality: Int. Union For. Res. Org. (IUFRO) workshop, New Zealand, August 1979. For. Chron. 56(3):116-120.

van den Driessche, R.

1969. Measurement of frost hardiness in two-year old Douglas-fir seedlings. Can J. Plant Sci. 49:159-172.

van den Driessche, R.

1971. Response of conifer seedlings to nitrate and ammonium sources of nitrogen. Plant and Soil 34:421-439.

van den Driessche, R.

1973a. Different effects of nitrate and ammonium forms of nitrogen on growth and photosynthesis of slash pine seedlings. Aust. For. 36(2):125-137.

van den Driessche, R.

1973b. Prediction of frost hardiness in Douglas-fir seedlings by measuring electrical impedance in stems at different frequencies. Can. J. For. Res. 3(2):256-264.

van den Driessche, R.

1976. Prediction of cold hardiness in Douglas-fir seedlings by index of injury and conductivity methods. Can. J. For. Res. 6(4):511-515.

van den Driessche, R.

1980. Effects of nitrogen and phosphorus fertilization on Douglas-fir nursery growth and survival after outplanting. *Can. J. For. Res.* 10:65-70.

Wakeley, Philip C.

1949. Physiological grades of southern pine nursery stock. *Proc. Soc. Am. For.* 1948:311-322.

Zaerr, J.B.

1972. Early detection of dead plant tissue. *Can. J. For. Res.* 2:105-110.