BOTRYTIS CINEREA PERS. ON CONIFER SEEDLINGS IN BRITISH COLUMBIA CONTAINER NURSERIES - A REVIEW

by

Jodi Ann Carlson

B.Sc. Simon Fraser University 1980

PROFESSIONAL PAPER SUBMITTED IN PARTIAL FULFILLMENT OF

THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF PEST MANAGEMENT

in the Department

of

BIOLOGICAL SCIENCES

SIMON FRASER UNIVERSITY

April 1986

Approval

Name: **Jodi Ann Carlson**

Degree: Master of Pest Management

Title of Professional Paper:

Botrytis cinerea Pers. on Conifer Seedlings in British Columbia. Container Nurseries - a Review.

Examining Committee:

Chairman:

Dr. J. E. Rahe, Professor, Senior Supervisor

Dr. J. H. Borden, Professor

Dr. J. R. Sutherland, Senior Research Scientist, Pacific Forestry Centre, Victoria, B.C.

Dr. G. R. Lister, Assistant Professor, Simon Fraser Univerity, Public Examiner

Date Approved: April 15, 1986

PARTIAL COPYRIGHT LICENSE

I hereby grant to Simon Fraser University the right to lend my thesis, project or extended essay (the title of which is shown below) to users of the Simon Fraser University Library, and to make partial or single copies only for such users or in response to a request from the library of any other university, or other educational institution, on its own behalf or for one of its users. I further agree that permission for multiple copying of this work for scholarly purposes may be granted by me or the Dean of Graduate Studies. It is understood that copying or publication of this work for financial gain shall not be allowed without my written permission.

Title of Thesis/Project/Extended Essay

Botrytis cinerea Pers. on Conifer Seedlings in British Columbia.

Container Nurseries - a Review.

Author: _____
(signature)

Jodi Ann Carlson
(name)

April 4, 1987
(date)

ABSTRACT

Grey mould of container-grown conifer seedlings in British Columbia forest nurseries is caused by Botrytis cinerea Pers. and affects foliage of seedlings during the growing season and in subsequent cold storage. Control of disease due to this fungus is becoming increasingly important as accelerating needs for reforestation cause a shift in nursery production methods from bareroot to container culture.

The objectives of this paper are to describe the circumstances leading to the B. cinerea moulding problem in B.C. via:

1. a review of seedling production techniques
2. an examination of host-pathogen interactions
3. a review of current control methods.

Based on findings in B.C. and elsewhere, recommendations are made for areas of investigation into control of this disease.

Prevention of disease requires an integrated approach with major emphasis on cultural methods to render the environment unsuitable for the pathogen. While timely applications of fungicides are likely beneficial, the potential for rapid build-up of resistant populations necessitates close monitoring of treatment efficacy.

Economic losses from disease in the nursery and following planting, due to effects on growth and yield and direct mortality, have not been investigated in B.C. Quantitative assessment of the problem is necessary before a mandate to reduce B. cinerea losses to a minimum can be received.

ACKNOWLEDGEMENTS

I would like to thank Drs. J. Rahe, J. Sutherland and J. Borden for assistance with this manuscript; Mr. W. Nijholt for translation of German literature; and Messrs. J. Sweeten, T. Willingdon and J. Halusiak for their time and donation of nursery stock.

TABLE OF CONTENTS

I. INTRODUCTION

Operational reforestation began in British Columbia (B.C.) in 1939, with approximately 4000 ha per year being planted between 1941 and 1965 (Johnson 1981). A paper presented at a 1974 symposium on seedling production reported that in Canada and the United States 50.6 million ha of productive forest land were in need of artificial regeneration and that current levels of reforestation were reducing the backlog by less than 2% annually (Bingham 1974). In B.C., with a 47.4 million ha public forest base (Anon. 1984a), annual reports of the B.C. Ministry of Forests indicate that on Crown Land in 1981 and 1982, 39% (Anon. 1983a) and 45% (Anon. 1984a) of the approximately 200,000 ha surveyed annually were classified as not satisfactorily restocked (NSR). To meet the reforestation needs under rapidly expanding harvesting rates in B.C. (Kinghorn 1970), container seedling culture was investigated as a method to supplement existing bareroot seedling production and make more effective use of manpower through increased mechanization. Kinghorn (1970) stated that rising labour costs and the scarcity of social capital available to forestry operations accentuated the urgency for developing planting methods with the capacity for huge gains in productivity.

Following several years of cultural experimentation (Matthews 1971) and planting trials (Vyse et al. 1971), a large scale container seedling program was established by the B.C. Ministry of Forests. In 1980, an inventory of public nurseries showed production of 33 million container seedlings, versus approximately 137 million bareroot seedlings (Anon. 1981a). The 1981-86 B.C. Ministry of Forests Five-Year Forest and Range Resource Program calls for production of 150 million plantable seedlings in 1985-86, with one-third of

these to be produced by private nurseries (Anon. 1981b). Of these seedlings, 50% will be container stock (Johnson 1981). To ensure adequate production of plantable seedlings in 1985-86, the report stated that a total of 168 million seedlings should be grown in that year to compensate for unavoidable annual losses (Anon. 1981b).

Some portion of the annual loss in nurseries is due to pest problems in the crop. While most pests encountered in container culture have been seen earlier in bareroot nurseries (Sutherland et al. 1981), some, particularly grey mould caused by the fungus Botrytis cinerea Pers., have assumed much greater importance. Peterson (1974) stated that Botrytis had caused more losses in containerized facilities than any other pathogen, resulting in >20% mortality of Douglas-fir (Pseudotsuga menziesii [Mirb.] Franco) seedlings in some greenhouses. Sutherland and Van Eerden (1980) noted that foliar diseases were of much greater importance on container-grown than bareroot stock and found that the incidence of B. cinerea had increased steadily with increasing container culture and was prevalent in nurseries throughout B.C. Infection or contamination of container stock by B. cinerea during the growing season leads to moulding in overwinter storage (Sutherland et al. 1981), thereby increasing losses to this pathogen.

The objectives of this paper are to describe the circumstances leading to the B. cinerea moulding problem in B.C. container nurseries, with particular attention to seedling production techniques and their development, an examination of Botrytis-conifer interactions and current control methods for this problem. Based on findings in B.C. and elsewhere, recommendations are made for investigations on control of this disease.

II. NURSERY PRACTICES

A. Container Culture

Container culture was initially investigated in B.C. as a method of decreasing reforestation costs through increased mechanization of the planting process (Kinghorn 1970). The first studies of container culture were in 1957, with seedlings grown in milk cartons (Johnson 1981), and by 1961 a method had been developed for growing and planting seedlings in individual plastic containers which became known as "Walters' bullets" (Walters 1961). With the use of a specially-developed planting gun, planting rates of about 2400 seedlings per person-day were achieved with "bullets" versus 400-900 for conventional bareroot stock (Cayford 1972). Although the "bullets" were structurally weakened and slit to facilitate root egress, field trials begun in 1967 showed that seedling survival was significantly improved if the container was removed prior to planting, allowing better root-soil contact (Van Eerden 1972; Arnott 1975). Frost heaving was also found to be a problem with seedlings planted in "bullets", whereas it was negligible for plugs, i.e., seedlings planted with containers removed (Van Eerden 1972).

The "Walters' bullet" had been designed for ease of planting, rather than rearing seedlings, and in 1967 a joint Canadian Forestry Service/B.C. Ministry of Forests project was initiated to develop a container which lent itself to mechanization of procedures in the nursery and from which seedlings would be removed at the planting site (Arnott 1973). A moulded styrofoam block was developed which had round, tapered cavities of approximately twice the volume of "bullets" (Cayford 1972), allowing firm, unbound root plugs to form which facilitated dibble planting (Kinghorn 1970). Rather than each cavity being an individual unit as with "bullets", each "BC/CFS Styroblock 2" had 192 cavities and could be loaded with growing medium, seeded and covered

with grit mechanically (Arnott 1973). An operational trial in 1970 showed that planting rates could be as high as 1200-1800 seedlings per person-day when seedlings were planted directly out of quartered styroblocks (Vyse et al. 1971), a considerable increase over bareroot planting rates.

The first large-scale container facility was constructed by the B.C. Ministry of Forests in 1971 at Surrey and had a capacity of about 8 million seedlings (Arnott 1973). In the same year, a publication was issued by the Canadian Forestry Service, Victoria, entitled "Container Seedling Production: a provisional manual" which detailed nursery construction and all phases of container seedling culture (Matthews 1971). Container facilities established by the B.C. Ministry of Forests over the next decade at new and existing nurseries numbered 10 by 1980, and produced a total of 31.7 million seedlings in that year (Anon. 1981a). A new Forest Act was proclaimed in 1978 which allowed production of forest seedlings by non-ministry nurseries (Johnson 1981).

The importance of production of healthy, adequately-sized stock was illustrated early in outplanting trials, where failures were attributed to low-quality seedlings. A strong impetus for container seedling production, especially in eastern Canada, was the potential for producing large numbers of seedlings in very short periods i.e., 8-12 weeks (Cayford 1972), which could be sown and outplanted continuously through the growing season (Kinghorn 1970). Field tests soon showed, however, that very small seedlings had poor survival rates. In Quebec in 1966-67 plantings were made of 8-week old black spruce (Picea mariana [Mill.] BSP) in plastic open-ended "Ontario tubes" when secondary growth was only 1.5-2.0 cm above the edge of the tubes (Arnott 1969). After one growing season, 55-60% of the seedlings were clas-

sified as dead/unhealthy (terminal dead or dying), of which 86% at one site had been killed by frost heaving or waterlogging.

A 1967 field trial compared survival of Douglas-fir and western hemlock (Tsuga heterophylla [Raf.] Sarg.) plug stock classed as small and regular (Arnott 1971). Average survival of small stock 3 years after planting was approximately 50% less than that of regular. Since the small seedlings were in poor condition at planting due to too much shade and no fertilizer during the growing season, this test likely reflected the need for healthy, vigourous seedlings at outplanting as much as a need for certain size criteria to be met (Arnott 1972).

Planting trials of Douglas-fir, lodgepole pine (Pinus contorta Dougl. ex Loud) and white spruce (Picea glauca [Moench] Voss) produced by various means showed that both size and age of stock at planting influenced seedling establishment (Van Eerden 1972). Van Eerden recommended that standards for size as well as quality be established for container-grown stock. High positive correlations between seedling height at outplanting and growth increment and height for several years after planting have been shown for southern pines (Barnett 1982); height at planting was the characteristic most indicative of subsequent field performance. Stein and Owston (1977) noted that while required seedling height may be attained 3 months after germination, additional growing time is required to achieve desired stem diameter, root mass and dormancy.

Although the use of styroblocks rather contradicted the original intent of container growth in B.C., i.e. mechanized planting, several advantages over bareroot seedling production have been realized from it. Nursery location is not restricted by physical factors important in bareroot nurseries, such as soil quality and depth of water table (Hahn 1982; Harris 1982), and

requires only a suitable climate and water supply (Kinghorn 1970).

Relatively small areas are required for production, with plant densities of approximately 6 million container seedlings/ha, versus 0.4-0.5 million/ha for bareroot stock (J. Halusiak[1], pers. comm.). Thus shelters for seedlings ranging from simple shadecloth over gravel or cement (Matthews 1971) to more elaborate fibreglass or glass houses can readily be constructed. These structures, depending on their complexity, allow the grower to manipulate light intensity, moisture, relative humidity and temperature (McDonald 1982). In southwestern B.C., where the growing season is long and the climate moderate, germination of seedlings is frequently carried out under controlled conditions with germinants later moved to open shadehouses (Matthews 1971). Supplemental lighting may be supplied to seedlings from high altitude or high latitude seed sources to provide the critical day lengths necessary to inhibit premature cessation of shoot growth and terminal bud formation (Arnott and Mitchell 1981; Arnott and Macey 1985). In areas with cooler climates supplemental heat, requiring enclosed structures (Stein and Owston 1977), may be needed at the beginning or end of the growing season.

Close control of cultural conditions can be maintained from sowing through to extraction (Hahn 1982), removing non-inherent variability within a crop which could occur in the field with bareroot seedlings. Uniform composition and volume of growing medium within a crop of container seedlings allow development of the "best" cultural conditions (Kinghorn 1970) which, in theory, should benefit all seedlings equally. These conditions contrast sharply with bareroot culture, where aspect, slope, soil composition and

---

1/ CIP Inc., Saanich Forestry Centre, Saanichton, B.C.

therefore, moisture and nutrition, can vary within a planting, resulting in a compromise in which only some portion of the crop is at optimum conditions.

In contrast to bareroot culture, applications of water, nutrients and pesticides to container seedlings are achieved with a minimum of labour via overhead irrigation systems, and minimal cost of materials due to the small areas and soil volumes involved (Matthews 1971). Since container seedlings are usually an annual crop, there is seldom any carryover of pest or nutritional problems (Matthews 1971). In addition, container culture, with its well-controlled germination and growth, is felt to make the most effective use of seed, especially if in limited quantity or genetically improved (Hahn 1982; Armson 1981).

B. Cold Storage

Storage practices for container stock are based on research with bareroot hardwood and conifer trees which has been conducted since the 1930's in the United States (Ruth 1953; Hopkins 1958). Extensive storage studies were begun in the 1950's in Europe (Sandvik 1959), England (Aldhous 1964), the US (Deffenbacher and Wright 1954; Simon 1961) and Canada (Jorgensen and Stanek 1962; Wilner and Vaartaja 1958). The objectives of storage have been many although the most important, from biological and operational points of view, was to provide stock for planting at sites climatically separated from the nursery by latitude or elevation, or both. Stored stock could be maintained in a dormant condition for late planting at high elevation long after the nursery climate had warmed (Simon 1961), and, conversely, trees could be removed from storage and planted before the climate in the nursery warmed sufficiently to allow lifting (Wilner and Vaartaja 1958; Eliason 1962).

Deffenbacher and Wright (1954) pointed out that storage allows quantity

production in lifting and packing, thereby maximizing use of manpower, and that planting contractors could order trees in small quantities, eliminating the need for heeling-in stock at planting sites until it could be used. Cold storage was investigated as a means to allow surplus seedlings from one year to be saved and used the next (Aldhous 1964). Mullin and Bunting (1972) stated that extension of the regular spring planting season was one of the chief advantages being sought in overwinter storage. More recently, Hinesley (1982) found that late fall lifting and storage of Fraser fir (Abies fraseri [Pursh.] Poir.) eliminated the problem of severe terminal bud abortion that accompanied spring lifting. Frost heaving is a serious problem with bareroot seedlings overwintered in the ground at the nursery; Johnson (1981) reported that in the period from 1975-1976 the B.C. Ministry of Forests lost 22 million 1+0 Interior spruce through frost heaving. Lifting and storage of seedlings in the fall avoids this problem. Cold overwinter storage also avoids the damage which can occur on overwintered container seedlings in cool greenhouses or shadehouses due to sudden freezes, desiccating winds and stimulation of foliar activity on sunny days (Stein and Owston 1977).

Packing methods in storage have included burying entire bundles of 50 or 100 seedlings in moist sand (Eliason 1962), layering seedlings on open trays with moist packing materials or upright in open crates with the roots in moist sphagnum moss or washed shingle tow (Deffenbacher and Wright 1954), and bundling seedlings tightly in bales of 750 with burlap or waterproof packing materials leaving bottom and top ends open (Mullin and Bunting 1972). The above methods, and especially the first, required large storage areas and it was found that open-stored seedlings were subject to desiccation (Deffenbacher and Wright 1954). Storage trials of seedlings in bundles or loose in

open or closed polyethylene or polyethylene-lined paper bags have shown varying degrees of success (Mullin and Bunting 1972; Bunting 1974), however, it appears that closed, poly-lined paper bags provide the best conditions for the seedlings in storage. In B.C., seedlings are stored in bundles in closed, poly-lined kraft paper bags inside waxed cardboard cartons at about 0°C (Hopkins 1975).

III. MOULDING BY BOTRYTIS CINEREA

A. The Fungus

i. Taxonomy

The genus Botrytis was erected by Micheli in 1729, making it one of the first genera of fungi described, and in 1801 Persoon designated five species including B. cinerea, which had been named by von Haller in 1771 (Jarvis 1977). Lack of a correct type species and perhaps misidentification of many fungi over the years resulted in 380 species being assigned to the genus by 1960 when Hennebert, in a doctoral thesis, re-evaluated the genus and reduced the number of species to 22 (Hennebert 1973).

The relationship of the imperfect (conidial) B. cinerea to a perfect (apothecial) stage has been the subject of numerous studies. The perfect stage of the fungus was thought to be Peziza fuckeliana by de Bary in 1864, although Fuckel transferred it to Sclerotinia in 1869 and Whetzel placed it in the genus Botryotinia in 1945 (Jarvis 1977). It was not until 1953 that a positive genetic connection was established between Botryotinia fuckeliana (de Bary) Whetz. and Botrytis cinerea Pers. ex Pers. (Groves and Loveland 1953). A 1974 Commonwealth Mycological Institute description lists B. cinerea as the conidial state of Sclerotinia fuckeliana (de Bary) Fuckel (Ellis and Waller 1974); later works place it in Botryotinia (Jarvis 1977, 1980a).

ii. Distribution and Host Range

B. cinerea occurs throughout the world and is most common in humid temperate or subtropical areas (Ellis and Waller 1974). A survey of the Review of Applied Mycology showed 235 host records for this fungus (MacFarlane 1968) and an annotated index of plant diseases and fungi lists 163 hosts in Canada, Alaska and Greenland (Conners 1967). Although primarily a pathogen of angiosperms (Jarvis 1977), B. cinerea has been recorded on non-vascular plants such as bryophytes (Anderson 1924), lower vascular plants including three genera of ferns (Anderson 1924) and gymnosperms (Peace 1962).

iii. Infection of Host

Infection by conidia is initiated through direct penetration of intact host cuticle with an infection peg from a germ tube or appressorium (McKeen 1974). Free water, present as a film or drop over the conidium, is necessary for germination to occur (Jarvis 1977). The length of time over which wetness is required to ensure infection varies with temperature. Nelson (1951), studying B. cinerea of table grapes, found that a wet period of 72-84 hours at 3°C was necessary for infection while a 12-24 hour period was sufficient at 16°C. Subsequent germ tube growth, appressorium formation and penetration may require only high relative humidity (Shoemaker and Lorbeer 1977; Jarvis 1980b). Whether penetration is via mechanical or enzymatic means, or a combination of both, has not been established (McKeen 1974; Jarvis 1977).

Once penetration is achieved, B. cinerea lives saprophytically, primarily in the host parenchyma, with hyphae advancing through tissue killed or dying from excreted fungal enzymes (Jarvis 1977). Conidia are borne in clusters on branched conidiophores which emerge from the infected tissue (Hennebert 1973).

Growth of B. cinerea mycelia occurs at temperatures as low as -2 to -3°C (Sommer et al. 1985) and as high as 35°C (van den Berg and Lentz 1968) with an optimum at 20-30°C, depending on the source consulted (Jarvis 1977). Van den Berg and Lentz (1968) found that growth was very slow at 0°C, with the rate increasing to a maximum at 25°C and dropping off rapidly at higher temperatures. Jarvis (1977) stated that the optimum temperature for growth was 22°C and that sporulation was greatest at 15°C. Germination of conidia has been recorded at temperatures from 0°C (Brooks and Cooley 1917) to 26°C (Jarvis 1977).

A study of the interaction of temperature and relative humidity (RH) on B. cinerea showed that mycelia survived, in the absence of nutrients, 2-12 months at 0°C and 85% RH and that survival was >12 months if the RH was raised to 95-100% (van den Berg and Lentz 1968). Conidia survived 2-6 months at 0°C and 85-99% RH, although an increase in temperature to 20°C with 85-90% RH reduced survival time to <1 month.

B. Epidemiology

i. Moulding During the Growing Season

Although grey mould of conifers is generally considered to be a nursery disease affecting bareroot (Halber 1963; Bloomberg 1966; James 1980) and container-grown seedlings (Sutherland et al. 1981; James et al. 1982), it has occasionally been found on older trees. Botrytis infection of young twigs of Norway spruce (Picea abies [L.] Karsten), Douglas-fir and European larch (Larix decidua Mill.) on forest trees of all ages was recorded in Germany following a spring of exceptional humidity (Zycha 1962). Peace (1962) noted that although Botrytis was uncommon on plantation conifers, appreciable damage had been found on succulent shoots of 2-3 m high Japanese larch (Larix

kaempferi [Lamb.] Carr.).

In the nursery, container-grown seedlings are much more likely than bareroot seedlings to show B. cinerea infection during the growing season (Sutherland et al. 1981) due to high crop density (Sutherland and Van Eerden 1980) and favourable conditions of moisture and humidity found in greenhouse/shelterhouse facilities (James and Woo 1984; Thies et al. 1980).

In contrast to perennial crops, such as strawberries, which carry inocula of sclerotia and mycelia over the winter (Jarvis 1962), annual infections in container-grown seedlings are due to airborne conidia (Sutherland et al. 1981) and possibly from contaminated irrigation water or seed (Sutherland and Van Eerden 1980). These early infections become established in senescent and damaged tissues (Jarvis 1977; Sutherland et al. 1981), since B. cinerea is basically a saprophyte (Jarvis 1977), and occasionally in young succulent growth (Jarvis 1980b; Cheung 1975). Mycelia are likely responsible for the plant to plant transmission of disease which leads to expanding infection centres within a crop. The saprophytic food base from which mycelia advance, plus the dependence of conidia on a film of free water for germination, give mycelia a much greater inoculum potential than that of germ tubes emerging from conidia (Jarvis 1977). Jarvis (1962) found in a field test that only 1.4% of infections of intact, ripe strawberry fruit could be attributed to conidia and that all others originated from mycelia in various saprophytic food bases. Following initial infection in a conifer seedling crop, the likelihood of disease establishment elsewhere within the crop is probably greatly increased by the large numbers of airborne conidia released from infection centres (Jarvis 1980b).

Botrytis epidemics are favoured by the cool, wet weather typical in B.C.

during the autumn and winter (Sutherland and Van Eerden 1980; Jarvis 1980b).

Fertilizers, especially nitrogen, can predispose seedlings to infection through stimulation of susceptible succulent growth (Jarvis 1980b) and from foliar burn due to application via overhead watering systems (Sutherland et al. 1981).

Many pesticides, including fungicides with *in vitro* activity against *B. cinerea*, have been reported to enhance epidemics of disease due to this fungus (Jarvis 1980b). This effect is likely due to phytotoxicity, either directly via injured tissue, or indirectly by causing a physiological change in the host. A release of nutrients to conidia from plants sprayed with ethylene-bis-dithiocarbamate fungicides was believed to enhance *B. cinerea* epidemics in tomatoes (Lockhart and Forsyth 1964). Douglas-fir seedlings treated with Bordeaux mixture showed 100% infection by *B. cinerea* while water treated control seedlings were disease-free (Bloomberg 1966). Bloomberg noted that the disease was stimulated by Bordeaux mixture and it seems likely that this occurred through predisposition of the host, possibly through the mechanism described by Lockhart and Forsyth (1964), rather than direct enhancement of the fungus.

Predisposition of the host due to frost damage has frequently been implicated in *B. cinerea* infections, especially on field-grown seedlings. Halber (1963) reported that *B. cinerea* had caused occasional, spotty infections of Douglas-fir seedlings in the nursery and felt that susceptibility of this species was governed by very specific factors, particularly unseasonable freezing weather. Peace (1962) reported that in some cases of *B. cinerea* infection on conifers, initial attack was on frosted tips of the seedlings.

In B.C. and the northwestern U.S., container-grown Douglas-fir and western hemlock are the species most frequently attacked by *B. cinerea* (Suth-

erland et al. 1981), but serious losses have occurred in lodgepole pine, ponderosa pine (Pinus ponderosa Laws.), Engelmann spruce (Picea engelmannii Parry) and western larch (Larix occidentalis Nutt.) in Montana and Idaho (James et al. 1982; James and Woo 1984). Grey mould is a common, serious disease on container-grown coast redwood (Sequoia sempervirens [D.Don.] Endl.) in northern California (McCain and Smith 1978).

Although susceptibility to infection varies among species of conifers, Peace (1962) stated that most species can likely become infected with B. cinerea under optimum conditions for fungal growth. He cited infections on Norway spruce, Scots pine (Pinus sylvestris L.) and European larch, all well-known for their resistance to this fungus. Differences in susceptibility between species have been attributed to many causes, both chemical and physical in nature.

A study of leaf waxes of Douglas-fir, Norway spruce and three species of pines showed that low concentrations of various cuticular wax fractions influenced germination and subsequent germ tube development of B. cinerea conidia in vitro (Schütt 1971). It was found that the stimulating effects of waxes from different species increased with the observed decreasing resistance of the species to infection; i.e. pine wax inhibited growth, spruce wax enhanced or did not affect growth and Douglas-fir wax always enhanced growth.

The small conidia of B. cinerea contain few reserves to ensure infectivity and exogenous nutrients stimulate germination, subsequent superficial mycelial growth and direct penetration or formation of appressoria (Jarvis 1980b). These nutrients occur in persistent moisture on leaf surfaces due to exosmosis of various sugars and acids by the host. Known to vary with host species (Kovacs and Szeoke 1956), the effects of these nutrients have not

been investigated for Botrytis-conifer interactions.

Growth habits of susceptible species such as Douglas-fir and western hemlock in containers lead to canopy closure by mid-summer and likely contribute directly to their high incidence of infection through reduced air flow and retention of moisture on the foliage (James and Gilligan 1983). Sutherland et al. (1981) stated that pine and spruce may be less affected by Botrytis due to their upright growth habits. Canopy closure not only creates a favourable microclimate for the fungus, but leads to senescence and abscission of lower non-photosynthesizing leaves (Sutherland et al. 1981), increasing the saprophytic food base.

ii. Moulding During Storage

Moulding of stock has been a common occurrence throughout the history of cold storage of conifers. Eliason (1962) stated that this was the only serious problem encountered in New York in numerous trials in several facilities and a review of trials in England showed that mortality of 10-40% of the seedlings in some tests was due to moulding (Aldhous 1964). This problem is considered to be of major significance in B.C. (Sutherland and Van Eerden 1980) where seedlings are routinely stored for periods up to 4.5 months (Hopkins 1975). Although disease occurrence varies from year to year, it can be common and severe in local nurseries. Extensive moulding was found on 6.5 million white spruce, western hemlock and Douglas-fir seedlings in B.C. in 1973-74 (Hopkins 1975). Bamford (1974) stated that the considerable trouble which was experienced in cold storage in 1973 was due to an outbreak of Botrytis which was aggravated by temperatures that reached 4.5°C in improperly designed facilities.

In contrast to the susceptibility differences seen during the growing

season, all species of container and bareroot seedlings grown in B.C. have shown some degree of moulding in storage (Sutherland and Van Eerden 1980). The first signs of disease are fluffy, grey mycelia on the lower foliage. This progresses upwards on the shoots and infected foliage becomes water-soaked in appearance, yellows and frequently abscisses (Sutherland and Van Eerden 1980). In severe cases, stem lesions and cambial death occur (Sutherland and Van Eerden 1980). Spread to adjacent seedlings in a bundle or to adjacent bundles is likely due primarily to mycelial growth in the same manner as during the growing season.

While storage moulding of container stock, with its soil-free growing medium, is due almost exclusively to B. cinerea (Sutherland and Van Eerden 1980), the growing environment of bareroot seedlings provides exposure to numerous soil-borne fungi which have been implicated as causal agents of storage moulding (Hopkins 1975). In local nurseries, isolations from moulded tissue have yielded Fusarium spp., Rhizopus spp., Penicillium spp. and many others (Sutherland and Van Eerden 1980). Fusarium spp., Trichoderma spp. and Penicillium spp. were identified as the fungi responsible for moulding in an early storage test of hardwoods (Wilner and Vaartaja 1958), while Hocking (1971) reported 37 species of fungi from moulded, cold-stored white spruce and lodgepole pine. Eliason (1962) stated that pathologists had identified the disease organisms only as fungi which grew well at near-freezing temperatures. Most storage trial literature reviewed for this paper referred to fungal infection or moulding without identifying the causal agents.

In B.C., moulding of stored container stock, especially of Douglas-fir and western hemlock, is felt to be a carryover from detectable B. cinerea infections established during the growing season (Sutherland et al. 1981). Storage moulding of the less susceptible container species and of bareroot

stock may be due to latent infections from the growing season (Sutherland et al. 1981) or to infections initiated in storage. Jarvis (1977) stated that most crops enter storage with incipient or latent infections of Botrytis incurred in the field, or are contaminated with conidia or saprophytic mycelia.

Predisposition of host tissues to infection in storage through decreased resistance potential at low temperatures has been found in various fruits, vegetables and woody materials (Jarvis 1977). Predisposition may be due to depletion of food reserves (Ritchie 1982), a factor implicated in the sudden onset of moulding in stored Sitka spruce (Picea sitchensis [Bong.] Carriere) (Buckley and Lovell 1974). Such predisposition may be important in both the transition from latent to active infections in stored seedlings and the establishment of new infections from conidia and mycelia.

IV. DISEASE CONTROL

A. Control During the Growing Season

1. Current Recommendations

Current recommendations for control of B. cinerea in container nurseries in the Pacific Northwest include cultural and chemical methods. Cooley (1981) stated that satisfactory control of Botrytis on conifer seedlings would never be achieved with fungicides alone, and that they must be used together with cultural controls. Lowering the relative humidity is recommended to make the environment less favourable for the fungus (Sutherland et al. 1981; Cooley 1981). This can be accomplished during the period of heaviest disease pressure, late summer onward, through spacing of containers, increasing air circulation and reducing irrigation, plus good sanitation

practices. Major control efforts, however, appear to rely on fungicides, with applications being made routinely via overhead boom or pressurized sprayers following canopy closure (Anon. 1984b; James and Gilligan 1983). Sutherland et al. (1981) suggested one or more applications of a systemic or protectant fungicide prior to canopy closure and alternating chemicals with different modes of action in subsequent sprays to avoid a build-up of resistant fungal populations. At present in B.C., and for the past several years, chlorothalonil, benomyl and captan are used for control of B. cinerea on seedlings in forest nurseries (Anon. 1981d; 1983b). Of these, only benomyl is registered for this use pattern, although chlorothalonil is registered for other diseases on nursery conifers and captan and captan/benomyl combinations are registered for control of Botrytis on other crops (Anon. 1981c; G. Shrimpton[2], pers. comm.).

Spray records from a large Vancouver Island nursery show eight applications of chlorothalonil from May 29 to December 21, 1984 with combination treatments of benomyl plus captan applied twice in November and once prior to extraction in January 1985 (Anon. 1984b). In the same nursery, nine applications of chlorothalonil were made between July 5 and November 10, 1983, with benomyl plus captan sprays in October 1983 and January 1984 (Anon. 1983b). Heavy reliance is being placed on a single chemical, chlorothalonil, for disease control throughout the growing season. As suggested by Sutherland et al. (1981), this repeated use can lead to rapid development of resistant fungus populations.

2/ Surrey Nursery, B.C. Ministry of Forests, Surrey, B.C.

ii. Fungicide Resistance

Resistance or tolerance of B. cinerea to fungicides has been reported in the literature for decades. The ability of the fungus to develop resistance in culture in the presence of chlorinated nitrobenzenes, such as PCNB and TCNB, was observed as early as 1947 (Jarvis 1977). Subsequent workers have found resistance to most fungicides in vitro, including copper and mercury salts (Parry and Wood 1958) and have demonstrated, through repeated subculturing, the ability of the fungus to grow at extremely high levels of these materials (Parry and Wood 1959). While these studies indicate what might occur during field use, close monitoring of development of resistance in the field likely provides a better measure of the efficaceous lifespan and hence, suitability for continued use, of a chemical. The case history of benomyl is particularly relevant to disease control efforts in B.C. container nurseries.

The benzimidazoles were introduced in the 1960's as the first fungicides showing "systemic" activity (Martin 1971) and benomyl, released by DuPont as Benlate® in 1968, was widely adopted for use in the control of diseases caused by B. cinerea and numerous other fungi (Thomson 1982). Investigations into the mechanisms of resistance in B. cinerea were stimulated following a report of acquired resistance to benomyl on cyclamen in a nursery in the Netherlands in 1971 (Bollen and Scholten 1971). Benomyl had been used only three times in this nursery, once in the previous year and twice on the current crop, when two sprays at 1.5-2.0 times the label rate not only failed to control the disease but led to unprecedented losses to Botrytis. The authors postulated that the chemical acted as a mutagenic agent on the fungus in the field. Polach and Molin (1975) later showed that resistant isolates of B. cinerea occurred at a frequency of 1 in 5×10^7 conidia in a sensitive

isolate derived from a single field-collected ascospore of Botryotinia fuckeliana. They suggested that, since this ascospore isolate had not been treated with mutagenic agents in the field, benomyl resistant mutants could arise spontaneously under field conditions. Polach and Molin, however, were unable to find such mutants in the field.

The first evidence of naturally occurring B. cinerea resistance to benomyl in the field was found in strains of the fungus on snapdragon plants which had never been exposed to fungicides (Bolton 1976). Conidia of a single isolate from these plants germinated and produced sporulating colonies on first exposure to 300 ppm of benomyl in an agar medium. While earlier workers had found cross-resistance to other benzimidazole fungicides including thiophanate-methyl, fuberidazole and thiabendazole (Bollen and Scholten 1971; Polach and Molin 1975), Bolton also found resistance in this isolate to two unrelated fungicides, triadimefon at 60 ppm and dicloran at 700 ppm. He postulated that "nuclei resistant to several fungicides may be present already in the cells of the fungus in nature and that combinations of resistant and susceptible nuclei exist in the spores, which, under selective pressure from these fungicides, reproduce rapidly."

The pathogenicity of fungicide resistant versus sensitive, wild-type strains of B. cinerea has been studied extensively. Bollen and Scholten (1971) reported that their benomyl-resistant isolates were less pathogenic and likely less competitive than sensitive isolates. Polach and Molin (1975) found that while a resistant isolate was somewhat less aggressive when inoculated onto bean leaves and pods, it sporulated more profusely than the sensitive isolate in the presence or absence of benomyl and showed comparable linear mycelial growth. High levels of benzimidazole-resistant strains of B. cinerea were isolated from vineyards where these compounds had not been

applied for the preceeding 4 years (Schuepp and Kung 1981), indicating that resistance did not confer a competitive disadvantage to these strains of the fungus.

Isolates of B. cinerea, from apple and strawberry fruits, showing stable resistance to captan were as pathogenic as non-resistant wild-types (Barak and Edgington 1984). Captan enhanced sporulation in 9 of 10 isolates of B. cinerea in vitro (James and Gilligan 1983) and low concentrations of both captan and thiram can stimulate germination of B. cinerea conidia on agar (Jarvis 1977). Some strains of B. cinerea tolerant to dicarboximide fungicides are less competitive than sensitive strains in vitro and in vivo (Gullino and Garibaldi 1981). It is evident that pathogenicity and competitive ability of fungicide resistant strains of B. cinerea vary with both the strain of the fungus and the chemical being tested.

iii. Fungicide Studies

The potential of various fungicides to control B. cinerea on container-grown seedlings has been studied extensively in vitro and in vivo. Much of the work has been done by the Forest Service in the western U.S.

a. Studies in vitro

Gillman and James (1978, 1980) collected isolates of B. cinerea from necrotic tissues of lodgepole and Scots pine and Engelmann and blue spruce (Picea pungens Engelm.) at three Colorado nurseries. Widespread mortality of lodgepole pine had occurred at one nursery despite fortnightly applications of benomyl, which had not been previously used at that facility. At a second nursery, where benomyl had been used for several years, mortality due to B. cinerea had occurred annually. Losses due to this fungus were found during the first year in a new greenhouse at a third nursery, again in spite of

routine benomyl applications. Radial mycelial growth of the fungus on potato dextrose agar amended with fungicides was used to evaluate tolerance of B. cinerea to the chemicals. Significant radial growth was observed in some or all of seven isolates tested on medium amended with 50 ppm of chlorothalonil, benomyl, captan, mancozeb or zineb, although the maximum growth on captan amended medium was only 16% of that observed on control medium. Tolerance of individual isolates to more than one fungicide was found. Dicloran was the only material tested for which no tolerance was observed in any of the seven isolates of the fungus, although other workers (Webster et al. 1970; Cooley 1981) have found tolerance to this chemical.

Ten isolates of Botrytis were collected from infected lodgepole pine at a Montana nusery where control failure with benomyl and chlorothalonil had occurred (James and Gilligan 1983). Growth of all isolates occurred on medium amended with 50 ppm benomyl or chlorothalonil. Nine of the 10 isolates grew on medium containing 50 ppm captan and significant growth was recorded for two isolates on 50 ppm vinclozolin. None of the isolates grew on plates containing 50 ppm iprodione or dicloran.

b. Studies in vivo

Early testing of the fungicide thiram (Volger 1959) indicated that it could be detected in conifer seedlings grown from seed which had been soaked in this chemical. Liquid extracts of these seedlings, with seedcoats removed, inhibited mycelia of B. cinerea on agar medium. This work stimulated studies in Oregon to assess the efficacy of thiram when applied to seedlings as a protectant against Botrytis (Halber 1963). Halber was successful in achieving infection of Douglas-fir seedlings only after subjecting them to temperatures of -5°C for 3-7 hours. He applied thiram as a slurry to these seedlings and

inoculated them with a suspension of B. cinerea conidia. The criterion for judging infection was apparently seedling death. While more seedlings died in treatments which were inoculated with the fungus than those which were not, neither this, nor a subsequent field trial performed on Douglas-fir seedlings which had been subjected to a sudden early frost, proved the activity of thiram against B. cinerea.

In the first apparent record of B. cinerea on conifer seedlings in a B.C. nursery, Bloomberg (1966) treated Douglas-fir seedlings with ferbam, thiram or zineb. Controls, sprayed with water only, and zineb-treated seedlings showed 73% and 56% infection by B. cinerea, respectively, while those treated with ferbam or thiram were disease-free. In additional testing he found that captan did not give complete protection from Botrytis.

McCain and Smith (1978) compared six fungicides for control of Botrytis on container seedlings of coast redwood. Fungicides were applied nine times at 14-day intervals throughout the growing season. Seedlings were inoculated by spraying with a suspension of Botrytis conidia between the first and second treatments. Disease was evaluated by counting lesions on the main stem 12 weeks after the first chemical treatment and 4 weeks following the final application. Dicloran and chlorothalonil provided excellent disease control at the first evaluation, with lesions on only 0.5% and 3.7% of seedlings, respectively. Lesions were observed on 27.5-38.5% of seedlings treated with mancozeb, captan, tri-basic copper and thiram and on 45.0% of the water-treated control seedlings. Four weeks following the last treatment, only 3.4% of the dicloran-treated seedlings showed lesions, significantly better than chlorothalonil at 51.0% and all other treatments at 82.5%-91.0%.

Six fungicides were evaluated as protectants against Botrytis on container-grown western larch seedlings in Idaho (James et al. 1982). Chemicals

were applied eight times at 14-day intervals throughout the growing season and seedlings were inoculated with a conidial suspension of B. cinerea after the second spray. Treatments were evaluated after the final spray, with disease being scored as present or absent. More than 50% of the seedlings treated with benomyl showed disease following the final treatment, significantly greater than captan at 29%. In contrast to the results of McCain and Smith (1978), dicloran was ineffective, with disease equal to that found on benomyl-treated seedlings. Nearly 100% of the water-treated control seedlings were diseased, while those in treatments of iprodione and two formulations of chlorothalonil showed less than 10% infection. Seedling survival was found to be directly related to the degree of disease control achieved by the treatments. Infection levels were too low to determine treatment effects in a duplicate test performed on bareroot western larch seedlings, despite two separate inoculations with B. cinerea (James and Woo 1984), and were also too low in a containerized western larch trial in Idaho (James and Genz 1983).

Thies et al. (1980) studied the effects of benomyl and captan on grey mould of Douglas-fir seedlings. Drenches of chemicals were made at sowing (pre-germination) or at appearance of true leaves (post-germination) or a combination of both on a 2-week re-treatment schedule. Seedlings in all treatments showed some level of disease at the termination of the test, approximately 6 months after sowing, with no significant differences between treatments and controls. The level of grey mould was also statistically equivalent to that found in treatments of ethazol and fenaminosulf, fungicides used as seed treatments or soil drenches for prevention of damping-off (Thomson 1982).

The results of these studies emphasize both the degree of resistance to all chemicals currently used in B.C. nurseries for control of B. cinerea and the variability between strains of the fungus. They also indicate that if resistance is present in high levels, even 2-week spray intervals will not provide adequate disease control.

c. Phytotoxicity

Along with poor efficacy due to fungal resistance and the potential for enhancing epidemics through predisposition of the host, fungicide treatments have frequently caused phytotoxicity to conifer seedlings. Wall (1974) cited excessive applications of fungicides as contributing to the death of more than 1 million conifer seedlings in the Maritime provinces from 1970-1974. Descriptions of damage range from slight chlorosis on shoot tips (James et al. 1982) to seedling defoliation and death (James and Genz 1983).

Captan has been known for many years to cause damage to seedlings. An inhibitory effect was seen on root and shoot growth of 5-6-week old Sitka spruce, Scots pine and western hemlock seedlings following a single application of 0.05% a.i. captan (Denne and Atkinson 1973), a level lower than the recommended rate for disease control (Anon. 1981c). Concomitant reductions in leaf size and weight occurred, becoming greater with increased concentrations and treatments. James et al. (1982) found significant reductions in height of container-grown lodgepole pine and western larch following multiple applications of captan at 2-week intervals using recommended rates.

Benomyl has reduced shoot growth of lodgepole pine (James et al. 1982) and Douglas-fir (Thies et al. 1980; Sutherland et al. 1977) although Sutherland et al. noted that, in general, benomyl was not phytotoxic to seedlings in the greenhouse or the field.

Treatment of container-grown western larch and lodgepole pine with

recommended rates of two formulations of chlorothalonil, i.e., Daconil® and Bravo®, plus dicloran and iprodione significantly decreased seedling height over controls (James et al. 1982), and Bravo® caused slight chlorosis of tips of western larch seedlings. These effects have not been noted in local nursery stock due to use of chlorothalonil.

Container-grown western larch treated with vinclozolin at label rates turned reddish-purple, defoliated prematurely and cambial tissues became necrotic, resulting in high mortality (James et al. 1982), while similar treatments of 2+0 bareroot stock at another nursery caused no damage (James and Woo 1984). James and Woo felt that this was not an anomaly, but that young, container-grown seedlings were more sensitive to treatment than older, field-grown stock. This effect had been previously described by Sutherland et al. (1977), studying the effects of single applications of various pesticides on greenhouse-grown (container) and field-grown (bareroot) coastal and interior provenances of Douglas-fir, Sitka and white spruce and lodgepole pine. They noted that in stock of comparable age, more phytotoxicity was observed in the greenhouse than in the field and that damage decreased in both as seedling age increased. Sutherland et al. (1977) pointed out that interactions with other pesticides, unfavourable weather and physiological condition of the seedlings at spraying may influence pesticide phytotoxicity.

B. Control During Storage

Current recommendations for control of storage moulding in B.C. nurseries emphasize prevention of disease prior to storage via effective disease control through the growing season and one or more applications of a systemic fungicide late in the growing season (Sutherland et al. 1981). These authors also suggest short storage periods, avoidance of foliar moisture when stock

is placed in storage and storage temperatures as low as stock can withstand.

It was recognized early in conifer storage testing that disease could be reduced through low foliar moisture and low storage temperatures. Deffenbacher and Wright (1954) found that if shoots of trees in adjacent, open-ended burlap bales touched, or if the moist sphagnum root packing material touched the foliage, moulding ensued. They recommended keeping as much of the tree shoots exposed as possible. Although desiccation of stock with this type of storage was a problem, temperatures of 0.5-1.0°C were low enough to suppress moulding, yet warm enough to allow maintenance of the 95% RH necessary to prevent excessive moisture loss.

Eliason (1962) felt that the most important way to reduce disease damage was to put trees into storage "field dry" and that if additional moisture was required it should be carefully applied to the roots only, or to the facility in general, to raise the humidity. This "field dry" recommendation, i.e. not adding moisture prior to storage, as was the practice, and ensuring that rain or dew from the field had evaporated, was also stressed by Aldhous (1964) who attributed moulding in several trials to packing trees when they were wet. Evidence in B.C. indicates that wetting foliage prior to storage may be beneficial in preventing moulding (Hopkins 1975; Sutherland and Van Eerden 1980), although current recommendations are to avoid storing wet stock (Sutherland et al. 1981).

Recommendations for sub-zero storage temperatures to control moulding were made as early as 1958, following very low outplanting survival of four hardwood species due to disease incurred in cold storage (Wilner and Vaartaja 1958). Subsequent work confirmed the efficacy of freezing temperatures for disease control (Mullin and Bunting 1972, 1979; Nyland 1974; Hinesley 1982;

Racey et al. 1984). Since the early 1960's there has been a shift from refrigerated to frozen overwinter storage, with temperatures of -3 to -5°C routinely being used in many northern areas, including Ontario (Mullin and Parker 1976; Wynia 1977), Sweden (Mattsson 1981) and the US Pacific Northwest (Ritchie 1984).

While trials comparing refrigerated and frozen overwinter storage for the past 25 years have yielded variable results, numerous tree species survive frozen storage well and when outplanted, frequently outperform refrigerated or fresh-lifted stock. This superior growth and survival has been attributed to many factors including lack of storage moulding (Hinesley 1982), either through direct suppression of the fungus (Sutherland and Van Eerden 1980) or through increased retention by the seedlings of stored metabolites, especially carbohydrates, for resumption of growth at planting (Mullin and Bunting 1972; Ritchie 1984), and to increased resistance to freezing weather after planting (Jorgensen and Stanek 1962).

Contradictory reports of seedling survival and vigour following frozen storage continue to appear in the literature. For instance, Cram and Lindquist (1981) found that storage at -5°C for 6.5 months was lethal to Scots pine, while Mattsson (1981) stated that in Sweden the normal temperature for an equivalent storage period is -5°C and that storage up to 9 months at these temperatures does not affect survival. Racey and Hutchinson (1983) found no difference in survival and performance between fresh or frozen Scots pine stock which had been stored 5 months at -3°C. Aldhous (1964) reported that Douglas-fir tolerated -5°C as well as +2°C for 5-6 months; longer storage at -5°C led to reduced survival. Ritchie (1984) stated that operational experience had shown that there was a clear preference for frozen storage of coastal provenances of Douglas-fir and other species for storage periods in

excess of two months. In contrast, van den Driessche (1977) found mean survival rates for coastal and interior provenances of Douglas-fir of 12% and 88%, respectively, following 4 months storage at -5°C and did not recommend frozen storage for this species.

The success of overwinter storage, generally measured as survival and growth of planted seedlings (Racey et al. 1984), depends heavily on the physiological condition of the seedlings when lifted. Seedling condition at lifting appears to be of even greater importance for frozen than for refrigerated stock (Hocking and Nyland 1971). The importance of lifting date was noted as early as 1958 when it was found that storage survival of Engelmann spruce and lodgepole pine progressively declined when trees were lifted at intervals following bud swell in the spring (Simon 1961). Likewise, Aldhous (1964) related some storage failures to lifting too late and stated that seedlings must be fully dormant when placed in storage. Lavender and Wareing (1972) found up to 80% mortality of early fall-lifted Douglas-fir following 4-6 weeks of storage at about 4°C, while seedlings lifted a few months later suffered no ill effects. Hocking and Nyland (1971), in a review of storage trials, suggested that stock was not fully dormant when performance of frozen seedlings was poorer than that of refrigerated stock, a view supported by Mullin and Parker (1976), who felt that losses during frozen storage may have been due to improper timing of the fall lift for storage.

To maximize storage survival, it has become evident that seedlings must be both cold-hardy and dormant (Hocking 1972; Tanaka 1974; Tinus 1981), and therefore in a state of maximum resistance to stress (Zaerr et al. 1981; Lavender and Stafford 1985) prior to storage and outplanting. While correct lifting date for bareroot seedlings exposed to natural environmental con-

ditions generally ensures that these requirements are met, container-grown seedlings take much longer than bareroot stock to achieve desired levels of hardiness (Timmis and Tanaka 1976) and artificial methods of increasing cold hardiness and inducing dormancy have been studied intensively.

These studies have focused on treatments during the latter part of the growing season and have included: withholding all nutrients (Etter 1972); withholding nutrients except potassium (Hocking 1972); withholding nitrogen and feeding with high levels of potassium and phosphorus (Tanaka 1974); warm, or cool, short day (8 h) photoperiods (Tanaka 1974; van den Driessche 1969); and moisture stress ranging from mild to severe (Timmis and Tanaka 1976; Blake et al. 1979). There do not yet appear to be standardized practices for dormancy induction in B.C. and current techniques encompass some aspects of all of the above (J. Halusiak, pers.comm.). Work with various growth retardants and inhibitors in western hemlock seedlings indicated that while four of five compounds tested caused some degree of phytotoxicity to the seedlings, these types of materials may have potential for use as dormancy inducers (Cheung 1975).

Several methods have been investigated for judging readiness of stock for storage. Bunting (1975) found a strong correlation between large stem diameter at lifting and survival at outplanting and recommended close adherence to culling standards based on diameters. He also proposed a Negative Degree Day system, based on cumulative nighttime air temperatures, whereby trees would not be lifted for storage until a predetermined threshhold was reached (Bunting 1975). The following year, the Ontario Ministry of Natural Resources published "Provisional guidelines for fall lifting for frozen overwinter storage of nursery stock". These guidelines were based on a similar concept, called Degree Hardening Days (Mullin and Parker 1976). Electrical

conductivity of stem leachates has also been used to measure the cold hardiness of seedlings (van den Driessche 1969).

Studies of frozen overwinter storage of seedlings have not been reported in B.C. and the technique does not appear to be routinely used for disease prevention (Sutherland et al. 1981). However, it is evident from the literature that several tree species grown here can withstand frozen storage, including white spruce (Jorgensen and Stanek 1962; Eliason 1962; Bunting 1970, 1974), Sitka spruce (Aldhous 1964), lodgepole pine (Aldhous 1964) and Douglas-fir (Aldhous 1964; Ritchie 1984). In addition, several species related to those grown here, including black spruce (Wynia 1977), European larch, Japanese larch and tamarack (Larix laricina [Du Roi] C. Koch) (Mullin and Lucas 1982), Fraser fir (Hinesley 1982) and numerous pines (Racey and Hutchinson 1983; Mullin and Parker 1976; Bunting 1975) have performed well in the field following extended frozen storage.

V. DISCUSSION

Research on grey mould of container-grown seedlings in B.C. has not been reported in the literature. The economic importance of the problem does not appear to have been evaluated and actual loss assessments are precluded by the current culling practices at extraction, which do not include a breakdown of the causes for which a seedling is considered unsuitable for storage or planting.

Little work has been reported on the effect of moulding on survival and growth of seedlings after planting. While field performance of lightly infected Scots pine and blue spruce was found to be unimpaired (Cram and Lindquist 1981), a study of moulded lodgepole pine reported decreased performance in the field (Hocking 1971). Studies of moulded, outplanted trees would

allow a threshold for a biologically acceptable degree of infection to be established for each species.

Methods of control of B. cinerea during the growing season must be, as Sutherland et al. (1981) recommended, a combination of cultural and chemical techniques. While spacing of containers during the latter part of the growing season likely reduces disease pressure through lowered humidity around the styroblocks, container facilities I have visited are filled to capacity to maximize production and, especially in greenhouses, there is little or no room to space the blocks out. Even if blocks are spaced, it does not alter the fact that within a 160 cavity styroblock seedlings are at a density of approximately 8 million/ha or $800/m^2$, versus bareroot seedlings at about $50/m^2$ in the field. While some nurseries are starting to use blocks with only 110 cavities, the cavity size will be 50% larger (J.Halusiak, pers. comm.), and it seems possible that some or all of the canopy density reductions which could be realized may be lost for some species due to increased seedling size. A reduction in cavity number per styroblock, with minimal or no increase in cavity size might have a significant effect in lowering the degree of canopy closure and making the environment less favourable for B. cinerea. Whether this wider spacing would decrease the number of seedlings produced to a greater extent than the losses due to disease could be investigated, during grading at extraction, in parallel tests of the systems.

While many contraindications for fungicide use on conifer seedlings have been presented it is unlikely that, even with good cultural practices, their use can be avoided completely. A rational approach to their use requires knowing their strengths and weaknesses.

Fungicide treatments should be monitored continuously in container seed-

ling nurseries for both short-term disease control and in the longer-term for development of resistance. Development of province-wide protocols for testing and reporting would provide reliable, standardized information upon which nursery pest managers could make control decisions and which would indicate to researchers the areas of study required. Guidelines for chemical use, particularly in the areas of application frequency and tactical choices for avoidance of resistance, should be established. Fungicide delivery systems could be evaluated to determine whether presently used equipment is the best available; recent studies indicate that fungicide applications via overhead irrigation systems are much less effective in depositing material on the foliage than are pressurized sprayers (Smith and McCain 1981).

In spite of good management practices during the growing season, some moulding will continue to occur during refrigerated overwinter storage. For most species grown in B.C., frozen storage appears to be the best solution to this problem, and reports in the literature concur that moulding is completely prevented at temperatures around -5°C. Perhaps some species grown in B.C. may not be able to withstand frozen storage and will have to continue to be stored at 0-1°C. Recent research on B. cinerea moulding of roses in refrigerated storage has shown that enhancement of atmospheric carbon dioxide has helped to reduce disease (Phillips et al. 1985) and may have some application to conifers in storage. Close adherence to standard refrigerated storage recommendations of low foliar moisture, good culling procedures of dead or diseased stock and careful monitoring of storage conditions will help to lessen the severity of disease in refrigerated storage.

Due to its prevalence and ecology, B. cinerea will continue to cause disease in container nurseries. It appears that effective practices are available for maintaining losses to grey mould at acceptable levels.

VI. LITERATURE CITED

1. Aldhous, J.R. 1964. Cold storage of forest nursery plants - an account of experiments and trials; 1958-63. Forestry 37:47-63.

2. Anderson, J.P. 1924. Botrytis cinerea in Alaska. Phytopathology 14:152-155.

3. Anon. 1981a. Ministry of Forests Annual Report, 1980. Hazeldine Press Ltd., Victoria, B.C. 60 pp.

4. Anon. 1981b. Ministry of Forests five-year forest and range resource program. Queen's Printer, Victoria, B.C. 17 pp.

5. Anon. 1981c. Compendium of pest control products registered in Canada - control of plant diseases. Agric. Canada Publication 1654 PD/81. Pesticides Division, Plant Products and Quarantine Directorate, Agric. Canada, Ottawa.

6. Anon. 1981d. Pacific Forest Products Ltd., B.C. Min. of Env. Pest Control Service Operations Record - 1981 Greenhouse Crop.

7. Anon. 1983a. Ministry of Forests, Annual Report, 1981-82. Queen's Printer, Victoria, B.C. 55 pp.

8. Anon. 1983b. Pacific Forest Products Ltd., B.C. Min. of Env. Pest Control Service Operations Record - 1983 Greenhouse Crop.

9. Anon. 1984a. Ministry of Forests Annual Report, 1982-83. Queen's Printer, Victoria, B.C. 112 pp.

10. Anon. 1984b. Pacific Forest Products Ltd., B.C. Min. of Env. Pest Control Service Operations Record - 1984 Greenhouse Crop.

11. Armson, K.A. 1981. Containerized seedlings and Canada's forest renewal. pp. 5-8 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

12. Arnott, J.T. 1969. Survival of bare-root and container-grown black spruce in Quebec. Dept. of Env. CFS Bi-monthly Res. Notes 25:16-17.

13. Arnott, J.T. 1971. Progress report on field performance of Douglas-fir and western hemlock container seedlings on Vancouver Island, British Columbia. Dept. of Env. CFS Info. Rep. BC-X-63. 81 pp.

14. Arnott, J.T. 1972. Influences affecting container seedling performance on Vancouver Island, British Columbia. pp. 84-91 In Waldron, R.M., ed. Proceedings of a Workshop on Container Planting in Canada. Dept. of Env. CFS Info. Rep. DPC-X-2. Ottawa. 168 pp.

15. Arnott, J.T. 1973. Evolution of the styroblock reforestation concept in British Columbia. Commonw. For. Rev. 52:72-78.

16. Arnott, J.T. 1975. Field performance of container-grown and bareroot trees in coastal British Columbia. Can. J. For. Res. 5:186-194.

17. Arnott, J.T. and D.E. Macey. 1985. Effects of supplemental light intensity on white spruce, Engelmann spruce and mountain hemlock seedlings grown under an extended photoperiod. Can. J. For. Res. 15:295-301.

18. Arnott, J.T. and A. Mitchell. 1981. Influence of extended photoperiod on growth of white and Engelmann spruce seedlings in coastal British Columbia nurseries. pp. 139-152 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

19. Bamford, A.H. 1974. Development of the British Columbia container program. pp. 53-60 In Tinus, R.W., W.I. Stein and W.E. Balmer, eds. Proceedings of the North American Containerized Forest Tree Seedling Symposium. Great Plains Agric. Council Pub. No.68. Denver, Colorado. 458 pp.

20. Barak, E. and L.V. Edgington. 1984. Botrytis cinerea resistant to captan: the effect of inoculum age and type on response to the fungicide. Can. J. Plant Path. 6:211-214.

21. Bingham, C.W. 1974. Achieving reforestation goals. pp.3-7 In Tinus, R.W., W.I. Stein and W.E. Balmer, eds. Proceedings of the North American Containerized Forest Tree Seedling Symposium. Great Plains Agric. Council Pub. No.68 Denver, Colorado. 458 pp.

22. Blake, J., J. Zaerr and S. Hee. 1979. Controlled moisture stress to improve cold hardiness and morphology of Douglas-fir seedlings. For. Sci. 25:576-582.

23. Bloomberg, W.J. 1966. Grey mould of Douglas-fir seedlings. Dept. of Env. CFS Bi-monthly Res. Notes 22:7.

24. Bollen, G.J. and G. Scholten. 1971. Acquired resistance to benomyl and some other systemic fungicides in a strain of Botrytis cinerea in cyclamen. Neth. J. Plant Path. 77:83-90.

25. Bolton, A.T. 1976. Fungicide resistance in Botrytis cinerea, the result of selective pressure on resistant strains already present in nature. Can. J. Plant Sci. 56:861-864.

26. Brooks, C. and J.S. Cooley. 1917. Temperature relations of apple-rot fungi. J. Agric. Res. 8:139-164.

27. Buckley, G.P. and P.H. Lovell. 1974. The effect of cold storage on subsequent growth of one-year-old seedlings of Picea sitchensis. Ann. Bot. 38:657-660.

28. Bunting, W.R. 1970. Preliminary report cold storage experiment Orono Nursery - 1968/69. Ontario Dept. of Lands and For. Nursery Notes No. 25. 5 pp.

29. Bunting, W.R. 1974. Cold storage tests. Ontario Min. of Nat. Res. Nursery Notes No. 37. 11 pp.

30. Bunting, W.R. 1975. More cold storage trials. Ontario Min. of Nat. Res. Nursery Notes No. 44. 9 pp.

31. Cayford, J.H. 1972. Container planting systems in Canada. For. Chron. 48:235-239.

32. Cheung, Kin-Wah. 1975. Induction of dormancy in container-grown western hemlock: effects of growth retardants and inhibitors. B.C. For. Serv. Res. Note No. 73. 9 pp.

33. Conners, I.L. 1967. An annotated index of plant diseases in Canada and fungi recorded on plants in Alaska, Canada and Greenland. Can. Dep. Agric. Res. Branch Pub. 1251. 381 pp.

34. Cooley, S.J. 1981. Fungicide tolerance of Botrytis cinerea isolates from conifer seedlings. USDA FS Pacific NW Region. 13 pp.

35. Cram, W.H. and C.H. Lindquist. 1981. Overwinter and spring storage of pine and spruce seedlings. For. Chron. 57:162-164.

36. Deffenbacher, F.W. and E. Wright. 1954. Refrigerated storage of conifer seedlings in the Pacific northwest. J. For. 52:936-938.

37. Denne, M.P. and L.D. Atkinson. 1973. A phytotoxic effect of captan on the growth of conifer seedlings. Forestry 46:49-53.

38. Eliason, E.J. 1962. Damage in overwinter storage checked by reduced moisture. Tree Planters' Notes 55:5-7.

39. Ellis, M.B. and J.M. Waller. 1974. Sclerotinia fuckeliana (conidial state: Botrytis cinerea). CMI Descriptions of Pathogenic Fungi and Bacteria No. 431. Commonwealth Mycological Institute. Kew, England.

40. Etter, H.M. 1972. Physiological basis for the manipulation of seedling characteristics. pp. 67-71 In Waldron, R.M., ed. Proceedings of a Workshop on Container Planting in Canada. Dept. of Env. CFS Info. Rep. DPC-X-2. 168 pp.

41. Gillman, L.S. and R.L. James. 1978. Fungicidal tolerance of Botrytis cinerea. USDA FS Rocky Mtn. Region State and Private For. Rep. R2-16. 15 pp.

42. Gillman, L.S. and R.L. James. 1980. Fungicidal tolerance of Botrytis within Colorado greenhouses. Tree Planters' Notes 31:25-28.

43. Groves, J.W. and C.A. Loveland. 1953. The connexion between Botryotinia fuckeliana and Botrytis cinerea. Mycologia 45:415-425.

44. Gullino, M.L. and A. Garibaldi. 1981. Competition in vitro and in vivo between strains of Botrytis cinerea Pers. sensitive and resistant to dicarboximides. Neth. J. Plant Path. 87:243.

45. Hahn, P.F. 1982. A historical overview of the use of containerized seedlings for operational reforestation. How did we get where we are today? pp. 7-12 In Guldin, R.W. and J.P. Barnett, eds. Proceedings of the Southern Containerized Forest Tree Seedling Conference. USDA FS Southern For. Exp. Sta. Gen. Tech. Rep. SO-37. New Orleans. 156 pp.

46. Halber, M. 1963. Botrytis sp. on Douglas fir seedlings. Plant Dis. Rep. 47:556.

47. Harris, H.G. 1982. Bare root versus containerized seedlings: a comparison of production problems and methods. pp. 77-80 In Guldin, R.W. and J.P. Barnett, eds. Proceedings of Southern Containerized Forest Tree Seedling Conference. USDA FS Southern For. Exp. Sta. Gen. Tech. Rep. SO-37. New Orleans. 156 pp.

48. Hennebert, G.L. 1973. Botrytis and Botrytis-like genera. Persoonia 7:183-204.

49. Hinesley, E.L. 1982. Cold storage of fraser fir seedlings. For. Sci. 28:772-776.

50. Hocking, D. 1971. Effect and characteristics of pathogens on foliage and buds of cold-stored white spruce and lodgepole pine seedlings. Can. J. For. Res. 1:208-215.

51. Hocking, D. 1972. Current rearing knowledge. pp. 48-66 In Waldron, R.M., ed. Proceedings of a Workshop on Container Planting in Canada. Dept of Env. CFS Info. Rep. DPC-X-2. 168 pp.

52. Hocking, D. and R.D. Nyland. 1971. Cold storage of coniferous seedlings - a review. State Univ. NY College of Env. Sci. and For. Syracuse, NY. Appl. For. Res. Inst. Res. Rep. 6. 70 pp.

53. Hopkins, G.M. 1958. Survival of nursery stock after cold storage. Fox For. Notes 11.

54. Hopkins, J.C. 1975. A review of the moulding of forest nursery seedlings in cold storage. Env. Can. CFS Info. Rep. BC-X-128. 19 pp.

55. James, R.L. 1980. Engelmann spruce needle blight at the Coeur d'Alene nursery, Idaho. USDA FS Northern Region State and Private For. Rep. 80-21. 5 pp.

56. James, R.L., J.Y. Woo and J.F. Myers. 1982. Evaluation of fungicides to control Botrytis blight of containerized western larch and lodgepole pine at the Coeur d'Alene nursery, Idaho. USDA FS Northern Region State and Private For. Rep. 82-17. 13 pp.

57. James, R.L. and D. Genz. 1983. Fungicide tests to control Botrytis blight of containerized western larch at the Champion Timberlands Nursery, Plains, Montana. USDA FS Northern Region Co-op. For. and Pest Man. Rep. 83-12. 7pp.

58. James, R.L. and C.J. Gilligan. 1983. Fungicidal tolerance of Botrytis cinerea from the Flathead Indian Reservation greenhouse, Ronan, Montana. USDA FS Northern Region State and Private For. Rep. 83-5. 15 pp.

59. James, R.L. and J.Y. Woo. 1984. Fungicide trials to control Botrytis blight at nurseries in Idaho and Montana. Tree Planters' Notes 35:16-19.

60. Jarvis, W.R. 1962. The epidemiology of Botrytis cinerea Pers. in strawberries. Proc. Int. Hort. Congress 1961:258-262.

61. Jarvis, W.R. 1977. Botryotinia and Botrytis species: taxonomy, physiology and pathogenicity. A guide to the literature. Can. Dept. Agric. Res. Branch Monograph No. 15. 195 pp.

62. Jarvis, W.R. 1980a. Taxonomy. pp. 1-18 In Coley-Smith, J.R., K. Verhoeff and W.R. Jarvis, eds. The Biology of Botrytis. Academic Press. NY. 318 pp.

63. Jarvis, W.R. 1980b. Epidemiology. pp. 219-250 In Coley-Smith. J.R., K. Verhoeff and W.R. Jarvis, eds. The Biology of Botrytis. Academic Press. NY. 318 pp.

64. Johnson, C.M. 1981. The status of container planting programs in Canada 1. British Columbia. pp. 19-21 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling symposium. COJFRC Symposium Proceedings O-P-10. 427 pp. NY. 318 pp.

65. Jorgensen, E. and W.K.L. Stanek. 1962. Over-winter storage of coniferous seedlings as a means of preventing late frost damage. For. Chron. 38:192-202.

66. Kinghorn, J.M. 1970. The status of container planting in western Canada. For. Chron. 46:466-469.

67. Kovacs, A. and E. Szeoke. 1956. The phytopathological implication of cuticular excretions [in German]. Phytopath. Z. 27:335-349.

68. Lavender, D.P. and S.G. Stafford. 1985. Douglas-fir seedlings: some factors affecting chilling requirement, bud activity, and new foliage production. Can. J. For. Res. 15:309-312.

69. Lavender, D.P. and P.F. Wareing. 1972. Effects of daylength and chilling on the responses of Douglas-fir (Pseudotsuga menziesii [Mirb.] Franco) seedlings to root damage and storage. New Phytol. 71:1055-1067.

70. Lockhart, C.L. and F.R. Forsyth. 1964. Influence of fungicides on the tomato and growth of Botrytis cinerea Pers. Nature 204: 1107-1108.

71. McCain, A.H. and P.C. Smith. 1978. Evaluation of fungicides for control of Botrytis blight of container-grown redwood seedlings. Tree Planters' Notes 29:12-13.

72. McDonald, S.E. 1982. Fully controlled or semi-controlled environment greenhouses - which is best? pp. 81-85 In Guldin, R.W. and J.P. Barnett, eds. Proceedings of the Southern Containerized Forest Tree Seedling Conference. USDA FS Southern For. Exp. Sta. Gen. Tech. Rep. SO-37. New Orleans. 156 pp.

73. MacFarlane, H.H. 1968. Plant host-pathogen index to volumes 1-40 (1922-1961), Review of Applied Mycology. Commonwealth Mycological Institute, Kew, England. 820 pp.

74. McKeen, W.E. 1974. Mode of penetration of epidermal cell walls of Vicia faba by Botrytis cinerea. Phytopathology 64:455.

75. Martin, H., ed. 1971. Pesticide Manual, 2nd edition. British Crop Protection Council. 495 pp.

76. Mathews, R.G. 1971. Container seedling production: a provisional manual. Dept. of Env. CFS Info. Rep. BC-X-58. 57 pp.

77. Mattsson, A. 1981. Cold storage of containerized planting stock and subsequent performance after outplanting. pp. 239-244 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

78. Mullin, R.E. and W.R. Bunting. 1972. Refrigerated overwinter storage of nursery stock. J. For. 70:354-358.

79. Mullin, R.E. and W.R. Bunting. 1979. Container and tying variations in frozen overwinter storage of red pine. Ontario Min. of Nat. Res. Nursery Notes No. 63. 4 pp.

80. Mullin, R.E. and J.S. Lucas. 1982. Overwinter storage tests of larch and tamarack. Ontario Min. of Nat. Res. Nursery Notes No. 76. 6 pp.

81. Mullin, R.E. and J.D. Parker. 1976. Provisional guidelines for fall lifting for frozen overwinter storage of nursery stock. For. Chron. 52:22-25.

82. Nelson, K.E. 1951. Factors influencing the infection of table grapes by Botrytis cinerea. Phytopathology 41:319-326.

83. Nyland, R.D. 1974. Sub-freezing temperatures control storage moulding. State Univ. N.Y. College of Env. Sci. and For., Syracuse, NY. Appl. For. Res. Inst. Res. Note 11. 3 pp.

84. Parry, K.E. and R.K.S. Wood. 1958. The adaptation of fungi to fungicides: adaptation to copper and mercury salts. Ann. Appl. Biol. 46:446-456.

85. Parry, K.E. and R.K.S. Wood. 1959. The adaptation of fungi to fungicides: adaptation to captan. Ann. Appl. Biol. 47:1-9.

86. Peace, T.R. 1962. Pathology of trees and shrubs - with special reference to Britain. Oxford at the Clarendon Press, London. 753 pp.

87. Peterson, G.W. 1974. Disease problems in the production of containerized forest tree seedlings in North America. pp. 170-172 In Tinus, R.W., ed. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

88. Phillips, D.J., D.A. Margosan and D.C. Fouse. 1985. Postharvest control of Botrytis rot of roses with carbon dioxide. Plant Dis. 69:789-790.

89. Polach, F.J. and W.T. Molin. 1975. Benzimidazole-resistant mutant derived from single ascospore culture of Botryotinia fuckeliana. Phytopathology 65:902-904.

90. Racey, G.D. and R.E. Hutchinson. 1983. Overwinter storage of Scotch pine at St. Williams nursery. Ontario Min. of Nat. Res. Nursery Notes No. 87. 3 pp.

91. Racey, G.D., R.E. Hutchison and C. Glerum. 1984. Effect of extended cold storage after overwinter frozen storage of white pine and white spruce. Ontario Min. of Nat. Res. Nursery Notes No. 105. 5 pp.

92. Ritchie, G.A. 1982. Carbohydrate reserves and root growth potential in Douglas-fir seedlings before and after cold storage. Can. J. For. Res. 12:905-912.

93. Ritchie, G.A. 1984. Effect of freezer storage on bud dormancy release in Douglas-fir seedlings. Can. J. For. Res. 14:186-190.

94. Ruth R.H. 1953. Survival and growth of fresh and stored planting stock. USDA FS Pacific NW For. and Range Expt. Sta. Res. Note No. 93.

95. Sandvik, M. 1959. Winter storage of planting stock in a shed or in cold storage. Norsk. Skogbr. 4:231-233.

96. Schuepp, H. and M. Kung. 1981. Stability of tolerance to MBC in populations of Botrytis cinerea in vineyards of northern and eastern Switzerland. Can. J. Plant Path. 3:180-181.

97. Schütt, P. 1971. The effect of cuticular waxes on the infection rate of pathogenic fungi. 1. Lophodermium pinastri and Botrytis cinerea. Eur. J. For. Path. 1:32-50.

98. Shoemaker, P.B. and J.W. Lorbeer. 1977. The role of dew and temperature in the epidemiology of Botrytis leaf blight of onion. Phytopathology 67:1267-1272.

99. Simon, C.L. 1961. Effects of lifting date, cold storage and grading on survival of some coniferous nursery stock. J. For. 59:449-450.

100. Smith, P.C. and A.H. McCain. 1981. Comparison of two systems of applying fungicides for the control of Botrytis blight of redwood seedlings. Tree Planters' Notes 32:32-33.

101. Sommer, N.F., J.R. Buchanan, R.J. Fortlage and B.E. Bearden. 1985. Relation of floral infection to Botrytis blossom-end rot of pears in storage. Plant Dis. 69:340-343.

102. Stein, W.I. and P.W. Owston. 1977. Containerized seedlings in western reforestation. J. For. 75:575-578.

103. Sutherland, J., T.A.D. Woods and S. Ilnytzky. 1977. Pesticide phytotoxicity studies with seeds, germinants and seedlings of British Columbia conifers. Env. Can. CFS Info. Rep. BC-X-165. 7 pp.

104. Sutherland, J.R. and E. Van Eerden. 1980. Diseases and insect pests in British Columbia forest nurseries. BCMF/CFS Joint Rep. No. 12. 55 pp.

105. Sutherland, J.R., W. Lock and L.E. Benson. 1981. Diseases and insects and their management in container nurseries. pp. 215-223 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

106. Tanaka, Y. 1974. Increasing cold hardiness of container-grown Douglas-fir seedlings. J. For. 72:349-352.

107. Thies, W.G., P.W. Owston and D.C. Hansen. 1980. Effects of four fungicides on survival and growth of containerized Douglas-fir seedlings: greenhouse performance. Can. J. For. Res. 10:423-425.

108. Thomson, W.T. 1982. Agricultural chemicals. Book IV - Fungicides. 1982-83 Revision. Thomson Publications, Fresno, Ca. 181 pp.

109. Timmis, R. and Y. Tanaka. 1976. Effects of container density and plant water stress on growth and cold hardiness of Douglas-fir seedlings. For. Sci. 22:167-172.

110. Tinus, R.W. 1981. Environmental control of seedling physiology. pp. 75-82 In Scarratt, J.B., C. Glerum and C.A. Plexman, eds. Proceedings of the Canadian Containerized Tree Seedling Symposium. COJFRC Symposium Proceedings O-P-10. 427 pp.

111. van den Berg, L. and C.P. Lentz. 1968. The effect of relative humidity and temperature on survival and growth of Botrytis cinerea and Sclerotinia sclerotiorum. Can. J. Bot. 46:1477-1481.

112. van den Driessche, R. 1969. Influence of moisture supply, temperature, and light on frost-hardiness changes in Douglas-fir seedlings. Can. J. Bot. 47:1765-1772.

113. van den Driessche, R. 1977. Survival of coastal and interior Douglas-fir seedlings after storage at different temperatures, and effectiveness of cold storage in satisfying chilling requirements. Can. J. For. Res. 7:125-131.

114. Van Eerden, E. 1972. Influences affecting container seedling performance near Prince George, British Columbia. pp. 92-100 In Waldron, R.M., ed. Proceedings of a Workshop on Container Planting in Canada. Dept. of Env. CFS Info. Rep. DPC-X-2. 168 pp.

115. Volger, C. 1959. A systemic effect of the fungicidal agent TMTD [in German]. Naturwissenschaften 46:148-149.

116. Vyse, A.H., G.A. Birchfield and E. Van Eerden. 1971. An operational trial of the styroblock reforestation system in British Columbia. Dept. of Env. CFS Info. Rep. BC-X-59. 33 pp.

117. Wall, R.E. 1974. Recent conifer disease problems in forest nurseries in the maritime provinces. Can. Plant Dis. Surv. 54:116-118.

118. Walters, J. 1961. The planting gun and bullet: a new tree planting technique. For. Chron. 37:94-95.

119. Webster, R.K., J.M. Ogawa and E. Bose. 1970. Tolerance of Botrytis cinerea to 2,6-dichloro-4-nitroaniline. Phytopathology 60:1489-1492.

120. Wilner, J. and O. Vaartaja. 1958. Prevention of injury to tree seedlings during cellar storage. For. Chron. 34:132-138.

122. Wynia, A. 1977. Frozen storage and winter grading of spruce planting stock can improve first year growth. Ontario Min. of Nat. Res. Nursery Notes No. 46. 6 pp.

123. Zaerr, J.B., B.D. Cleary and J.L. Jenkinson. 1981. Scheduling irrigation to induce seedling dormancy. pp. 74-79 In Proceedings of Intermountain Nurseryman's Association and Western Forest Nursery Association Combined Meeting. USDA FS Intermountain For. and Range Exp. Sta. Gen. Tech. Rep. INT-109, Ogden, Utah. 148 pp.

124. Zycha, H. 1962. Botrytis damage to conifers [in German]. Phytopath. Z. 43:234-247.