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The relationship of site factors and the incidence of cytospora and septoria cankers and poplar and willow borer in hybrid poplar plantations in Mason and Manistee counties, Michigan

> Abebe, Gashawbeza, Ph.D. Michigan State University, 1988



THE RELATIONSHIP OF SITE FACTORS AND THE INCIDENCE OF CYTOSPORA AND SEPTORIA CANKERS AND POPLAR AND WILLOW BORER IN HYBRID POPLAR PLANTATIONS IN MASON AND MANISTEE COUNTIES, MICHIGAN.

By

Gashawbeza Abebe

A DISSERTATION

Submitted to

Michigan State University in partial fulfillment of the requirements for the degree of

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Department of Botany and Plant Pathology

1988

ABSTRACT

THE RELATIONSHIP OF SITE FACTORS AND THE INCIDENCE OF CYTOSPORA AND SEPTORIA CANKERS AND POPLAR AND WILLOW BORER IN HYBRID POPLAR PLANTATIONS IN MASON AND MANISTEE COUNTIES, MICHIGAN.

By

Gashawbeza Abebe

Two hybrid poplar plantations in the northwestern part of Lower Michigan were surveyed (1984-87) for the incidence of poplar and willow borer damage, and Cytospora and Septoria cankers. Tree heights, root starch levels, leaf water potentials, root development, disease incidence and clonal differences were measured to determine stand conditions. Soil samples were analyzed for physical and chemical properties.

At the Manistee site Cytospora canker was prevalent, while at the Mason site, Septoria canker, and poplar and willow borer were predominant. The incidence of Septoria canker was highly correlated to tree height and borer damage.

A discontinuous ortstein layer was present at both study sites. Eventhough there was no significant difference for the beginning of the ortstein layer; the maximum depth and thickness of the ortstein layer were significantly different between the two study sites. Tree root development and stem height were significantly reduced by the presence of the

ortstein layer.

Except for N and Ca, the two study sites were different in their soil nutrient contents. The Mason site had more P, K and Mg than the Manistee site, while the reverse was true for Fe, Mn and Al. Soil nutrients and tree height were the two factors which correlated most closely with the pest problems in the two plantations. Cytospora canker increased as Fe and Al levels increased and as K levels decreased. Septoria canker incidence, however, increased with increasing levels of P, K and Mg, and as Fe and Al levels decreased. Poplar and willow borer damage was found to increase with increasing P and K levels.

Using discriminant and stepwise regression analyses, equations accounting for more than 80% of the variation in the incidence of cankers and borer were developed. An interaction model between pest incidence, and soil and stand factors was developed.

DEDICATION

This dissertation is dedicated to my son Abiye G. Abebe; my sister Jerusalem K. Afework and her husband Hailu Afework, the two persons who have always been supportive of my education. Whatever I achieve in my life I owe it to you. May God bless you.

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GENERAL INTRODUCTION

Study Sites Description

The study sites were located in Manistee (Maple Grove Township, Section 29, T23N, R14W) and Mason (Custer Township, Section 3, T18N, R16W) counties in Michigan, on plantations established by Packaging Corporation of America (PCA). Between 1969 and 1981 PCA established about 1,000 hectares of hybrid poplar plantations in the northwestern part of Lower Michigan for its mill near Filer City. The mill makes a corrugated medium pulp and paper board.

The Manistee plantation was established in the spring of The plantation was a mixed stand of two hybrid poplar clones: NE 47 and NE 235. NE 47 [P. maximowiczii X (P. X berolinesis) cv. Oxford] is a cross between the Japanese poplar and the natural hybrid Berlin poplar (P. laurifolia X P. nigra). NE 235 (P. deltoides X P. nigra Incrassata) is a hybrid between eastern cottonwood and black poplar (Dickmann and Stuart, 1983; Woods, 1984). Previous to planting the site was an abandoned field. Before planting, the land was plowed and cuttings were machine planted with 2.4 by 3 m spacing. Herbicides were applied the second year for weed control. This study site was about 1.2 hectares and was initially planted with about 1605 cuttings in 23 rows. The soil was somewhat poorly to poorly drained, and the land had 0 to 2% slope. The water table was within 1.25 m from the surface and soil profiles were highly mottled. A cemented layer of

ortstein was sometimes observed. The plow layer was sandy and the soil taxon is Aeric haplaquod of the Finch soil series (Woods, 1984).

The Mason plantation (1.8 hectares) was established in the spring of 1979 with a mixture of cuttings of clones NE 47, NE 235 and NE 308. NE 308 (P. nigra var Charkowiensis x P. nigra Incrassata) is a cross between two black poplar varieties (Ministry of Natural Resources, 1983). The field was under cultivaton prior to planting with hybrid poplars. The site was plowed and herbicides applied before cuttings were machine planted with an average spacing of 2.4 by 3 m. Additional herbicides were applied the second year for weed control. Originally 2655 cuttings in 34 rows were planted. The soil was moderately well drained and the land had 0 to 2% slope. A discontinuous cemented layer of ortstein was observed. The plow layer was sandy loam and the soil taxon is Typic haplaquod of the Ogemaw series.

In 1984 both plantations had conspicuous gaps and open spaces where the trees had died.

Poplars

The genus <u>Populus</u> is in the Salicaceae family and consists of nearly 30 species, which are widely distributed in the Northern Hemisphere (Rehder, 1954; Dickmann and Stuart, 1983). Poplar culture began in the United States in 1784 when Lombardy poplar, <u>Populus nigra</u> L. cv. "Italica" was first introduced into the country (Rehder, 1954). Since then poplars have been planted for shelter belts,

ornamentals and pulpwood production. The first artificial hybridization of poplars in the United States was done in the spring of 1924 (Stout, Mckee and Schreiner, 1927; Stout and Schreiner, 1933).

Since the early 1950's the genus <u>Populus</u> has been the major single source of pulpwood in the Lake States (French, 1976). Intensive poplar plantations were being developed in the North Central Region of the U.S. to produce wood biomass in order to meet future demands for fiber and energy (Ostry, 1981). Poplars were also widely planted for sheltering orchards, fields and farms in Michigan and the Great Plains (Walters, et al., 1982) and Prairie Provinces of Canada (Hocking, 1970).

Populus hybrids are potentially useful trees in a short rotation fiber production system. Their ease of planting, low initial mortality, rapid diameter and height growth, and high weight yields are a few of their advantages (Bowersox and Ward, 1976). Lees and Anderson (1980) further suggested that the genus <u>Populus</u> might offer more possibilities for genetic improvement than any other genus of forest trees. However, to utilize their full potential, proper clone and site selections are very important. If planted off site the trees are susceptible to a number of pests and the expected yields are not achieved. This phenomenon was observed in the Packaging Corporation of America (PCA) hybrid poplar plantations in northwestern Lower Michigan, where growth rate was reduced, mortality was

high, and canker diseases and insect borers were prevalent. Poplars require high levels of moisture and nutrients and hence their growth is affected by site factors. Soil depth, soil fertility, soil pH, soil moisture and soil aeration are some of the site factors that should be considered before poplar plantation establishment (Dickmann and Stuart, 1983). The presence and size of sweet fern (Comptonia peregina) and bracken fern (Pteridium aquilinum) are good indicators of site quality for poplar (Graham, Harrison and Westell, 1963). Areas with a heavy growth of reeds (Phragmites communis), reed grass (Phalaris arundinacea) or sedges (Carex paludosa) are unsuitable (Peace, 1952). Generally the best sites are those with deep, medium textured soil where root penetration is not interrupted by bedrock, hardpans or gravel layers (Dickmann and Stuart, 1983; Gt. Brit. For. Commission, 1923). Areas with an impermeable layer within 30 cm of the surface are poor sites as a stagnant water table may develop (Graham et al., 1963). addition, when surface layers of such areas become dry, there is no opportunity for water replacement from below or for the roots to penetrate deep into moisture bearing strata.

OBJECTIVES

The objectives of this study were:

To determine if the incidence of Septoria canker,
 Cytospora canker and poplar and willow borer damage was influenced by site factors.

- 2. To determine if there was a spatial relationship between soil physical and chemical characteristics and the incidence of cankers in the hybrid poplar stands.
- 3. Based on the above relationships, to develop the "best" predictive equation(s) for Septoria canker, Cytospora canker, and poplar and willow borer damage to the three Populus hybrid clones in the study sites.

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THE INCIDENCE OF SEPTORIA CANKER, CYTOSPORA CANKER AND POPLAR AND WILLOW BORER IN TWO HYBRID POPLAR PLANTATIONS IN NORTHWESTERN MICHIGAN.

INTRODUCTION

Cytospora canker

Over 50 Cytospora species were reported on hardwood hosts in North America (Spielman, 1985). Cytospora chrysosperma (Pers.)Fr. (teleomorph: Valsa sordida Nits.) is one of the most common bark inhabiting fungi found on indigenous and introduced Populus species (Christensen, 1940; Hinds, 1976). It was first described by Long from southwestern U.S. in 1918 (Schreiner, 1931).

Cytospora canker on poplars caused by <u>C</u>. <u>chrysosperma</u> is widely distributed in the temperate zone of all continents except Africa (Browne, 1968). It is widely distributed in the U.S.A. In a disease study conducted in Alaska, Cytospora canker was one of the common diseases of aspen (Hinds and Laurent, 1978). In Wyoming, the fungus was isolated frequently in aspen stands (Ross, 1975) and in a forest nursery in North Dakota, <u>C</u>. <u>chrysosperma</u> destroyed more than 50% of the production of salable plants (Walters et al., 1982).

Cytospora canker is generally distinguished by black pycnidia in the dead bark of cankered stems. The fungus exudes allanoid, hyaline conidia in yellow to red mucilaginous tendrils during moist weather (Hepting, 1971). Biggs and Davis (1983) reported that colonization after inoculation occurs by mechanical ramification of dense wedges of large hyphae in the periderm, cortex and phloem tissues. The cells behind are colonized both inter- and

intra-cellularly by smaller hyphae. The cankers are long and narrow and the inner bark rapidly turns dark as the underlying sapwood is stained light brown. Generally dead bark remains attached to the tree for two to three years and then falls off in large pieces exposing the sapwood. Girdled or partially girdled stems often put out vigorous sprouts below the canker. Other symptoms include dead cambium with discolored, watery and smelly wood. (Hepting, 1971). According to Bloomberg (1962), canker development increased proportionally with temperature and inversely with soil moisture content. Filer (1967) reported that the optimum temperature for growth of the fungus as 25°C.

C.chrysosperma is not an aggresive parasite and attacks trees when their resistance is lowered by adverse conditions like severe pruning, unfavorable environment, fire injury (Povah, 1922), other diseases (Hocking, 1970), winter injury (Kuntz and Ricker, 1949) or insects (Anderson, Ostry and Anderson, 1979). Many researchers including Broadfoot and Farmer (1969), Schoeneweiss (1975, 1978, 1981), Bertrand et al., (1976), Bloomberg (1962), Bloomberg and Farris (1963), Christensen (1940), Filer (1967), Juzwick, Nishijma and Hinds (1978), Graham et al. (1963), Hinds (1976), Biggs and Davis (1981, 1983), Zsuffa (1975), Moss (1922) and Hepting (1971) have reported that trees need to be predisposed before attack by C. chrysosperma.

Septoria canker

Septoria canker is caused by the fungus <u>Septoria musiva</u>

Peck (teleomorph: <u>Mycosphaerella populorum</u> G.E. Thompson).

The fungus is indigenous to North America (Waterman, 1946).

It was first described by C.H. Peck in 1882 from <u>Populus</u>

<u>deltoides</u> leaves at Albany, New York (Peck, 1884).

- S. musiva produces leaf spots, branch and stem cankers of both native and introduced poplars (Palmer, Schipper and Ostry, 1980). Moore and Wilson (1983) considered S. musiva as the worst pathogen of poplars in nurseries and plantations in Michigan and Wisconsin. Septoria cankers are necrotic zones of depressed, cracked and blackened tissues (Long, Bowersox and Merril, 1986), which can be flat faced or can have marginal callus (Walla and Conway, 1986). dieback and breakage are commonly observed in poplar stands infected with S. musiva. The incidence and severity of Septoria canker varies by location within a region, and by clone, tree and seed sources (Zalasky, 1978; Ostry and McNabb, Jr., 1985). Hybrid poplar clones derived from P. trichocarpa, P. laurifolia or P. maximowiczii are very susceptible to S. musiva (Ostry and McNabb, Jr., 1986; Walla and Conway, 1986). Stems with cankers had a higher wood density compared to non-cankered stems (McNabb Jr., 1981), but the wood produced low quality fiber for pulping as it contained more lignin than healthy wood (Ostry and McNabb, Jr., 1983).
- S. musiva overwinters in fallen infected leaves and in stem and branch cankers. In spring ascospores and conidia

from fallen leaves and conidia from stem cankers are released during wet weather. Spores are spread by wind or washed by rain to infect stems and leaves. Infection occurs through stipules, petioles, buds and borer wounds. Secondary infections are caused by conidia from pycnidia from leaf spots. Warm temperatures and long periods of high humidity favor disease development (Riffle and Wysong, 1986).

Conidia are hyaline, cylinderic, straight or curved with one to four septa and are 20-50 µm by 3-4 µm in size. Ascospores are hyaline with one septum and are 16-28 um long and 4.5-6.0 um wide (Riffle and Wysong, 1986; Spielman, Hubbes and Lin, 1986; Waterman, 1954; Thompson, 1941).

Poplar and willow borer

Poplar and willow borer (Cryptorhynchus lapathi (L.), (Coleoptera: Curculionidae) attacks willows, poplars, alder and birch (Harris and Goppel, 1967; Matheson, 1917). It is native to Europe and Asia but was introduced to North America in New York City in 1882 (Furniss, 1972). It is now well distributed throughout southern Canada and the northern half of the United States (Harris and Goppel, 1967). Poplar and willow borer is also an important pest to poplars in Europe. It is reported as the most serious insect pest of poplar plantations in Quebec Province of Canada and in the Lake States of the U.S. by Morris (1981) and Moore and Wilson (1983) respectively.

The poplar and willow borer takes one to three years to complete its life cycle depending on latitude. In France it takes one year while in British Columbia, Canada, three years are needed to complete its life cycle (Harris and Goppel, 1967). In northwest Michigan <u>C.lapathi</u> takes one to two years to complete the life cycle depending on local weather conditions (Moore, 1984).

Adult poplar and willow borer weevils feed on young, succulent one year old shoots. After mating adult females cut small holes at lenticels, branch bases or at the edge of damaged bark. One egg is usually oviposited and covered with fine wood particles. Hatching occurs 18-21 days later and the larvae begin enlarging their holes immediately. Fine brown chips ejected from the holes by the larvae are the first evidence of damage to poplar trees. Initially, mining and feeding are usually round the trunk or branch. Later, larvae start mining inwards and upwards towards the heartwood of the tree and more frass is pushed out through small openings as the larvae grow. After pupation emerging adults leave the mines through the openings made by the larvae. The adult is black and white or grayish, robust, oval and about 1 cm long. Although winged, adults usually do not fly long distances (Moore, 1984; Matheson, 1917; Doom, 1966).

MATERIALS AND METHODS

Tree mortality

Tree mortalities from planting (1978 or 1979) to 1984, from 1984 to 1986 and during the summer of 1987 were calculated based on the original planting space of 2.4 X 3 m. Open spaces and dead snags were combined to tally for dead trees.

Tree height

Height of every tree at both locations was measured in the summer of 1984 using a telescopic measuring pole. Each tree was identified by clone either as NE 47, NE 235 or NE 308. A t-test was conducted to determine if there was a significant difference in mean of tree heights by study site.

Disease rating

In the summer of 1984 individual trees at both study sites were checked for the presence or absence of Cytospora canker. In the summer of 1986 trees were individually examined and rated for Septoria canker and poplar and willow borer in addition to Cytospora canker. In the 1986 observation both cankers and borer damage were rated using the following scoring scheme:

Rating	<u>Septoria and</u> Cytospora Cankers	Poplar and willow borer
1	Absence of canker	No apparent injury
2	Cankers on branches	Frass on the lower 1/3 of tree
3	Cankers on main stems	Frass on the lower 2/3 of tree
4	Main stem dieback, tree dead or dying	Frass on the lower 2/3 and the upper 1/3 of tree

Chi-square (X^2) values were calculated for cankers and borer rating classes to check for independence of site or clone. Further analysis of variance was carried out to determine clone and site interaction.

RESULTS AND DISCUSSION

Tree mortality

Between 1978-1984, 44% of the trees died at the Manistee plantation while 16% died at the Mason Plantation (Table 1). A second inventory in the summer of 1986 showed an additional mortality of 8% and 3% since the summer of 1984 at the Manistee and Mason study sites, respectively. By the summer of 1987, 60% at Manistee and 23% at Mason of the original planted trees were dead. In a Chi-square test comparison of tree mortality by year and study sites showed a significant interaction (P < 0.05).

Table 1. Summary of mortality of trees by study sites.

	ì	Mason	Ma	anistee
	No.of trees dead	% Mortality of original cuttings planted	No.of trees dead	% Mortality of original cuttings planted
Dead by 1984	415	15.6	705	43.9
Dead by 1986	495	18.6	837	52.1
Dead by 1987	611	23.0	968	60.3

The original planting at the Mason site was a mixture of clones, where the proportions or the total number of cuttings planted for each clone was unknown. Thus, it was not possible to determine mortality rates by clone prior to the 1984 inventory. However, between 1984 and 1987 at the Mason site mortality was 14%, 9% and 7% for clones NE 47, NE 235, and NE 308, respectively based on 1984 live trees of each clone (Table 2). Similar data were not available from the Manistee site as 98% of the trees were NE 47. However, NE 47 showed 29% mortality in 1987 based on 1984 residual live trees. These results suggest that location influenced mortality; the Mason site was a better site in terms of survival than the Manistee site.

Table 2. Summary of clone mortality between 1984 and 1987

		Mason	n		Manist	:ee
		live t			live tre	•
Clone	1984	1987	mortality	1984	1987	mortality
NE 47	288	248	13.9	884	623	29.5
NE 235	944	862	8.7	16	14	12.5
NE 308	1008	933	7.4	NOT	PLANTED)
Total	2240	2044	8.8	900	637	29.2

Tree height

Average tree heights for the three clones at the two study sites are shown in Table 3. Although the Mason plantation was one year younger, its trees were taller than those of the Manistee plantation. A t-test of average tree height in 1984 by location showed that trees at the Mason site were significantly taller (4.6 m) than trees at the Manistee site (3.1 m) (P < 0.01). In a clonal study in Pennsylvania and Maryland, NE 308 was ranked in the upper 12.5 percentile group after four years growth (Demeritt Jr.,1981). In this study NE 235 grew better than NE 308 at the Mason plantation.

Table 3. Tree heights in 1984 by clone and study site

Clone	Site	No. of trees	Ave. ht. in m	Std.error
	Mason	288	4.5	0.1
NE 47	Manistee	884	3.1	0.1
	Mason	944	4.9	0.1
NE 235	Manistee	16	4.2	0.3
NE OOG	Mason	1008	4.4	0.1
NE 308	Manistee	NOT	PLANTED	

Disease rating

Since all three clones were not planted at both study sites it was not possible to compare the effect of site on all clones. However, since NE 47 was planted at both study sites it was used as a basis to check if there was any difference in pest incidences between the two study sites (Table 4).

At both study sites Cytospora canker was more prevalent on branches than on the main stem of NE 47. However, more stem diebacks (27%) were observed on the trees at Manistee as compared to Mason (2%) (Table 4). A Chi-square test showed significant difference of Cytospora canker incidence between the two study sites (P < 0.01). Trees at Manistee were growing slower (Table 3), were more stressed than trees

Table 4. Comparison of cankers and borer ratings of NE 47 between the Mason and Manistee plantations.

Poplar and willow borer			Cyt	ospora	canker	Septoria	canker
	Mason	Maniste	e Mas	on	Manistee	Mason	Manistee
Rating*	No.of trees	No.of trees		.of ees %	No.of trees %	No.of trees %	No.of trees %
1	194 78	2 608 9	8.1 7	6 30.6	62 10.0	13 5.2	602 97.2
2	19 7	7 7 :	1.1 16	8 67.7	393 63.3	8 3.2	9 1.5
3	33 13	.3 3	0.5	0.0	1 0.2	16 6.5	6 1.0
4	2 0	8 2	0.3	4 1.6	164 26.5	211 85.1	2 0.3

* <u>Rating</u>	<u>Septoria and</u> Cytospora Cankers	Poplar and willow borer
1	Absence of canker	No apparent injury
2	Cankers on branches	Frass on the lower 1/3 of tree
3	Cankers on main stems	Frass on the lower 2/3 of tree
4	Main stem dieback, tree dead or dying	Frass on the lower 2/3 and the upper 1/3 of tree

at the Mason site, and hence were more suceptible to \underline{C} . chrysosperma.

Septoria canker on NE 47 was more prevalent at the Mason site than at the Manistee site. At the Manistee site only 3% of the trees showed symptoms of Septoria canker while at Mason 95% of the trees had symptoms by the summer of 1987 (Table 4).

Poplar and willow borer damage on NE 47 was more severe at the Mason stand (22%) than at Manistee (2%) (Table 4). A Chi-square test showed significant difference (P < 0.01) in poplar and willow borer damage between the two study sites.

At Mason 22%, 68% and 97% of the NE 47, NE 308 and NE 235 trees, respectively, were attacked by the borer. In Ontario, Canada-three-year old NE 47 plantations showed no infestation by poplar and willow borer, while NE 308 was infested 31 to 51 % (Morris,1981). Septoria canker and poplar and willow borer damage to NE 235 and NE 308 at the Mason plantation were very closely related (Table 5). The insect wound was probably serving as a good infection point for Septoria musiva (Moore, 1984).

At Mason absence of apparent Cytospora canker injury varied by clone; 31%, 88% and 98% of NE 47, NE 235 and NE 308, respectively, were free of Cytospora canker symptoms (Table 5).

At Mason 81% of NE 308 trees had Septoria canker as compared to 95% and 97% for NE 47 and NE 235, respectively. Stem canker and main stem dieback was observed in 92% of NE

47 trees, 94% of NE 235 trees and 70% of NE 308 trees. Unlike Cytospora canker, however, Septoria canker was found on the main stem as well as on the branches. Ostry (1987) reported that many hybrid poplar plantations in the North Central United States have failed due to S. musiva. Generally, severity of cankers, especially of Septoria canker at Mason, increased during the study period. Among the three clones at Mason, the fastest growing NE 235 was attacked more by S. musiva than NE 47 or NE 308. As S. musiva is an aggressive pathogen (Bier, 1939) it probably preferred the fast growing trees (Table 3) at Mason. In addition the wounds caused by the poplar and willow borer were serving as infection points for S. musiva.

The canker and borer ratings at the Manistee plantation are shown in Table 6. Since NE 235 comprised less than 2% of the plantation, ratings for NE 47 only are reported. At the Manistee plantation Cytospora canker was more prevalent than Septoria canker and poplar willow borer damage. The slow growing (Table 3) and stressed trees at the Manistee plantation were more susceptible to the nonaggressive facultative parasite Cytospora chrysosperma than to the aggressive Septoria musiva which prefers vigorously growing trees. The stressing site factors which predisposed the trees are discussed in the last part of this study.

As in other places where hybrid poplars are intensively planted, pathogens and insects were severely limiting the quality and productivity of the plantations.

TABLE 5. Canker and borer rating by clone in 1986 at the Mason plantation.

Poplar and willow borer						
	NE	47	N	235	NE	308
Borer rating	No.d			of es %	No.c	
1	194	78.2	26	3.0	298	31.9
2	19	7.7	36	4.2	97	10.4
3	33	13.3	770	89.3	374	40.1
4	2	0.8	30	3.5	164	17.6
Total	248	100.0	862	100.0	933	100.0

Cytospora canker

	NE 4	7	NE :	235	NE 3	308
	Canker No.of rating trees %		No.of trees %		No.of trees %	
1 2 3 4 Total	76 168 0 4 248	30.6 67.7 0.0 1.6 99.9	754 102 2 4	87.5 11.8 0.2 0.5	913 18 1 1 933	97.9 1.9 0.1 0.1
Iotai	240	39.3	002	100.0	333	100.0

TABLE 5. (cont'd).

Septoria canker

	NE 47		NE 235		NE 308	
Canker rating	No.of trees %		No.of trees %		No.of trees %	
1 2 3 4	13 8 16 211	5.2 3.2 6.5 85.1	24 23 34 781	2.8 2.7 3.9 90.6	179 105 575 74	19.2 11.3 61.6 7.9
Total	248	100.0	862	100.0	933	100.0

* <u>Rating</u>	<u>Septoria and</u> Cytospora Cankers	Poplar and willow borer
1	Absence of canker	No apparent injury
2	Cankers on branches	Frass on the lower 1/3 of tree
3	Cankers on main stems	Frass on the lower 2/3 of tree
4	Main stem dieback, tree dead or dying	Frass on the lower 2/3 and the upper 1/3 of tree

TABLE 6. Canker and borer rating in 1986 of NE 47 at the Manistee plantation

Poplar and willow bo			Cytosp canker		Septoria canker		
Pest	No.of		No.of		No.of	_	
rating	trees	*	trees	*	trees	s %	
1	608	98.1	62	10.0	602	97.2	
2	7	1.1	393	63.3	9	1.5	
3	3	0.5	1	0.2	6	1.0	
4	2	0.3	164	26.5	2	0.3	
Total	620	100.0	620	100.0	619	100.0	

* Rating	<u>Septoria and</u> Cytospora Cankers	Poplar and willow borer
1	Absence of canker	No apparent injury
2	Cankers on branches	Frass on the lower
		1/3 of tree
3	Cankers on main stems	Frass on the lower
		2/3 of tree
4	Main stem dieback,	Frass on the lower
	tree dead or dying	2/3 and the upper
		1/3 of tree

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TREE VIGOR AND PLANTATION CONDITION

INTRODUCTION

Root starch level

Tree vigor is defined as "an individual tree's physiological condition that results from its physiological performance within a particular environment" (Wargo, 1978). Tree vigor partially determines a tree's tolerance to stresses or attacks by insects and pathogens. One way of measuring tree vigor is to determine the starch content in the roots. In most deciduous trees starch is stored in high concentrations in the roots. Thus, root starch is the major carbohydrate reserve and reflects the photosynthetic capacity of the plant. Wargo (1976) reported that starch content in the roots was related to root diameter and varied by season. The maximum root starch level in sugar maple was in late fall, with a minimum in spring (Wargo, 1971). Starch levels in roots are either determined by chemical analysis (Hassid and Neufeld, 1964) or are estimated histochemically (Wargo, 1975). These two methods showed similar results in root starch content of black oak, white oak and sugar maple. Thus, Wargo (1981) reported that the histochemical method is faster and gives an equally accurate estimate of starch levels in deciduous tree roots.

Leaf_water potential

The water balance in plants influences the initiation and development of diseases (Schoeneweiss, 1978). Drought or water stress is considered as the major factor in predisposing blue spruce (Schoeneweiss, 1983; Kamiri and Laemmlen, 1981), poplars (Schoeneweiss, 1978) and French prune (Bertrand et al., 1976) to Cytospora canker.

Although the results are contradictory, a number of experiments have been conducted on how moisture stress predisposes trees to Cytospora canker. Bier (1964) suggested that these contradictions could be due to differences in methodology, inoculum potential or time of inoculation. To minimize these contradictions and for consistent results, Kramer (1963) suggested that drought should be measured in terms of water stress in the plant rather than in terms of soil water stress.

Plant water balance in poplar generally varies diurnally, seasonally, and by clone (Pallardy and Kozlowski, 1981). Water potential normally reached a minimum late in the morning. Slow growing <u>Populus</u> clones showed longer periods of low leaf water potential compared to that of fast growing clones. In another study (Biggs and Davis, 1981), the size of Cytospora cankers on hybrid poplar was positively correlated with bark relative turgidity and negatively correlated with bark water potential. Spruce stems subjected to -20 to -30 bars, developed cankers

whereas no cankers were observed on the controls at -2.5 bars (Schoeneweiss, 1983). Filer (1967) reported that July inoculations with <u>C</u>. <u>chrysosperma</u> produced significantly more stem cankers among 25 cottonwood clones than November inoculations, demonstrating the importance of temperature and moisture. Thus, drought stress could be the main predisposing factor for disease development in both hybrid poplars and in spruce.

Although many studies have been conducted on the effects of drought or water stress on woody species, comparatively few studies have been conducted on the effect of excess soil moisture. Excess soil water may be a problem on certain sites in Michigan (Robertson et al., 1979a). The type and level of injury caused by excess water differs with species, soil type and flooding time. Symptoms of injury include decreased growth rate of shoot and root, increased transpiration, leaf chlorosis, leaf epinasty, abscission, death of roots and increased susceptibility to attack by predators and pathogens (Gill, 1970; Kramer, 1940). Excess water predisposes trees directly by the exclusion of oxygen from the root system or by CO2 accumulation, and indirectly by accumulation of toxic products (Gill, 1970; Broadfoot, 1967). Most root diseases are favored by wet soils but according to Schoeneweiss (1975) the role of host predisposition in the majority of cases is not known. Robertson et al. (1979a), however, reported that adequately drained soil contains more air than poorly drained soil. Oxygen, the important component of air

in the soil, diffuses approximately 10,000 times faster through large well drained pore spaces in the soil than through water logged pore spaces. Robertson, Kidder and Mokma (1979b) further recommended surface drainage in order to reduce the level of soluble metals such as iron and manganese which are toxic in poorly drained and, especially, acidic soils. Season of flooding is important in predisposing trees to pathogens. Generally winter flooding is of little significance while excess water during the growing season is detrimental (Hall and Smith, 1955).

MATERIALS AND METHODS

Root starch level

On October 25, 1986; 54 roots were collected for root starch analysis. At the Manistee site 14 roots from 7 trees of NE 47 were sampled. At the Mason site 40 roots (20 roots from 10 trees of NE 235 and 20 roots from 10 trees of NE 308) were collected. Root samples were taken from trees nearest to the points where soil samples for nutrient analysis were taken (Figures 1 and 2). Root samples were about one cm in diameter and 30 cm in length. They were placed in a polyethylene bag and kept on ice for transporting to East Lansing. The samples were then kept at -20°C until they were analyzed for starch. Starch level was determined using potassium iodide (KI) solution following the procedure described by Wargo (1975)(Appendix A). After the staining, each cross section was examined under a light microscope at

100X magnification and photographed using Ektachrome color film. Roots were rated as depleted, low or medium in starch level based on the relative amount of staining.

Leaf water potentials

On June 17, 18 and July 29, 30, 1986 leaf water potentials (xylem water potential) of 62 trees were measured at the Mason plantation, using a Scholander's pressure bomb (Tyree and Hammel, 1972; Scholander et al., 1965; Tyree, Dainty and Hunter, 1974; Waring and Cleary, 1967; Ritchie and Hinckley, 1971). The 62 trees were tagged during the June measurements, hence the same trees were measured in July. Two small branches (5 to 10 mm in diameter) of approximately equal size from the same tree were taken 1.5 to 1.7 m above ground from the unshaded side of the tree. Measurements were taken between 10 A.M. and 1 P.M. with sunny skies. A weather station at Ludington, located about 16 km southwest of the plantation, recorded a minimum temperature of 6°C and a maximum temperature of 23°C between June 14 and 18. The minimum and maximum temperatures recorded for the July 26 to 29 period were 14°C and 29°C respectively. No precipitation was recorded for three days before or while leaf water potential measurements were taken on June 17 and 18. A total of 9 mm of rainfall was recorded for the three days (July 26, 27 and 28) before leaf water potential measurements were taken. However, there was no precipitation on July 29 and 30. Clone NE 235 and NE 308 were sampled equally throughout the stand. These trees were

randomly sampled from the better microsites (where trees were growing vigorosly) and stressing microsites (where trees were short and open spaces were present in the stand). Leaf water potential mean differences for NE 235 and NE 308 were compared by month of measurements and microsites using a t - test.

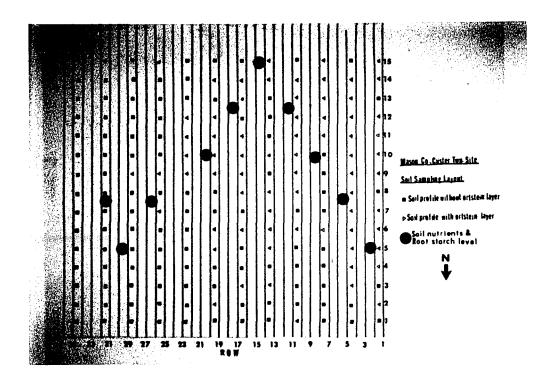


Figure 1. Field layout of samples taken for ortstein layer, soil nutrients and roots at Mason.

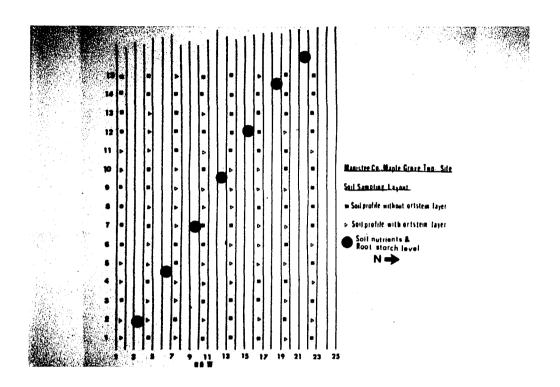


Figure 2. Field layout of samples taken for ortstein layer, soil nutrients and roots at Manistee.

RESULTS AND DISCUSSION

Root starch levels

In general root starch levels as measured with KI solution were low. Consequently, starch levels were grouped as depleted (0 to 1%), low (2 to 5%) or medium (6 to 10%). Root starch levels by clone and by study sites are shown in Table 7. When tree heights were compared by starch level groupings, a one way analysis of variance showed no significant difference in mean tree heights with starch levels (P > 0.05). A Chi-square test showed no significant differences when starch levels were compared by study sites (P > 0.05) or between clones (P > 0.05). Furthermore, a Chi-square test between starch levels and ratings for Septoria canker (P > 0.05) or Cytospora canker (P > 0.05) and poplar and willow borer damage (P > 0.05) showed no significant differences at Mason.

Leaf water potentials

Summary of leaf water potential means are shown in Table 8. No significant differences were observed between NE 235 and NE 308 trees on measurements taken on June 17 and 18 (P > 0.05) or July 29 and 30 (P > 0.05). Within the stand, when leaf water potential means of NE 235 and NE 308 were compared separately or together by microsites, no significant differences were observed at the 5% level of probability.

Table 7. Root starch level ratings summary

	Mani	Manistee*		Mason**				
	NE	47	NE	235	NE	308	TOT	AL
Starch level	n	*	n	%	n	%	n	%
Depleted (0-1%)	0	0	3	30	4	40	7	26
Low (2-5%)	5	71	7	70	5	50	17	63
Medium (6-10%)	2	29	0	0	1	10	3	11

^{*} as 98% of the stand was NE 47 the other clones were not found at the sampling points

Table 8. Leaf water potentials means in - bars at Mason plantation

	June	17 & 18,	1986	July	29 & 30,	1986
		Stressi micro- site	ng Total	Better micro- site	micro-	Total
Clone	n lwp	n lwp	n lwp	n lwp	n lwp	n lwp
NE 235	20 8.5	12 8.6	32 8.5	20 10.4	12 11.4	32 10.8
NE 308	19 10.7	12 9.5	31 10.2	19 12.4	11 12.2	30 12.3
Total	39 9.6	24 9.1	63 9.3	39 11.4	23 11.8	62 11.5

n = no. of trees

^{**} NE 47 was not sampled at this site

n = number of trees

lwp = leaf water potential

Plantation vigor could be estimated by combining the ratings of the sample trees in the stand (Wargo, 1978). Thus, the overall low root starch levels (2 to 5% staining of root cross-section with potassium iodide solution) indicated that trees in both plantations were not physiologically vigorous and probably were under stress. Lack of differences in pests incidence or tree height among root starch level groupings suggested that the trees had similar vigor.

For Botryosphaeria dothidea, a common pathogen which forms stem cankers on a wide range of woody plants predisposed by stress, the leaf water potential threshold canker development -12 to -13 for was (Schoeneweiss, 1981). In French prune orchards canker sizes caused by Cytospora leucostoma increased with increased water stress (Bertrand et al., 1976). The lack of significant differences in leaf water potential means between NE 235 and NE 308 indicated that both clones were similarly stressed at Mason.

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SOIL CHARACTERISTICS AND THEIR RELATIONSHIP WITH THE INCIDENCE OF SEPTORIA CANKER, CYTOSPORA CANKER AND POPLAR AND WILLOW BORER

INTRODUCTION

Stress factors and predisposition to pests

In general most species of higher plants are either immune or resistant to attack by the majority of microorganisms with which they come in contact (Schoeneweiss, 1975). Microorganisms enter resistant and susceptible hosts with equal frequency. The development of a disease condition, however, depends on the influence of environmental factors on the genetically controlled response of the host plant to the pathogen or its metabolites (Schoeneweiss, 1975).

Plant pathologists have long recognized the influence of environment on the epidemiology of numerous diseases (Schoeneweiss, 1981). But with the advent and wide use of fungicides in disease control, the importance of environmental stress and predisposition received less attention. Now, however, with public concern over environmental contamination and restrictions on the availability and use of fungicides, along with the development of the Integrated Pest Management (IPM) concept, there is a renewed interest in the study of environmental stresses as predisposing factors in disease development.

Environmental stress is defined as, "any factor capable of producing a potentially injurious strain; stress exerts the most pronounced effect in predisposing plants towards greater susceptibility to facultative parasites, particularly weak or non-aggressive parasites"

(Schoeneweiss, 1975). Nonaggressive facultative parasites like Cytospora chrysosperma, commonly enter host plants but remain latent or non-pathogenic until the host is stressed. Predisposition is defined by Schoeneweiss (1975) as "the tendency of non-genetic factors, acting prior to infection, to affect the susceptibility of plants to disease". Thus, predisposition implies an effect on the host rather than on the pathogen. Proneness or disposition of the host to disease prior to infection may influence the subsequent establishment and development of a pathogen (Schoeneweiss, 1978).

Generally, resistance to a given plant disease is a stable character and has its basis in the genetic composition of the host plant. But the expression of the genes may be modified by environmental factors (Barnett, 1959). These environmental factors (water stress, excess water, soil conditions, temperature, etc.) may modify resistance either by direct effect on the pathogen or indirectly through host metabolism.

<u>C. chrysosperma</u> attacks weakened plants or those predisposed by stress as it is a non-aggressive pathogen. It enters plants through wounds but does not cause disease damage as long as the host vigor is high (Schoeneweiss, 1981). On the contrary, <u>Septoria musiva</u> is an aggressive pathogen (Bier, 1939) which causes premature defoliation, reduces growth and predisposes susceptible trees to other pathogens and environmental stresses (Ostry and McNabb, Jr. 1983). <u>Cryptorhyncus lapathi</u>, a stem boring insect,

however, prefers trees that are weakened or are under stress (Wilson, 1976).

Soil characteristics and hybrid poplars

The soil environment is a major factor in determining whether trees survive and grow or whether they are suppressed and die. Roots respond to changes in the physical and chemical properties of soil depending upon the tree species and soil type (Rurark, Mader and Tattar, 1982). Soil chemical characteristics, e.g. nutrient levels, pH, cation exchange capacity (CEC), etc., and physical characteristics like soil texture, structure, drainage, compaction, etc., influence tree vigor and the productivity of the site. Unfortunately, there is lack of detailed soil chemical and physical characteristics data related to forest productivity as compared to agricultural lands. (1984) reported detailed soil descriptions and classifications of the PCA hybrid poplar sites. Yet, the report contained limited information on the chemical composition of the soil.

In establishing hybrid poplar plantations, Dickmann and Stuart (1983) suggested that soil fertility, pH, depth, moisture content and aeration be considered. Furthermore, due to the moderately deep rooting system of hybrid poplars, the nutrient content of the upper B horizon should be considered in addition to the A layer in site selections for hybrid poplar plantations (Roberts and Khalil, 1980). In a

study of soil nutrients in hybrid poplar plantations in central Pennsylvania; Ca, N and P were found to be important factors influencing tree height and stem wood yield (Bowersox and Ward, 1977). Studies with birch seedlings revealed that N, K and P levels decreased gradually with increasing uptake of Al while levels of Ca and Mg decreased rapidly, showing differential nutrient uptake response by the seedlings to Al level (Goransson and Eldhuest, 1987). Poplar plantation sites in Mason county, Michigan were found to be very deficient in P by Woods et al. (1982).

Hybrid poplar clones respond differently to nutrient levels. Clones derived entirely from crosses between P. deltoides and P. nigra were found to be more sensitive to Al than clones derived in part from P. trichocarpa, P. maximowiczii, P. balsamifera or P. laurifolia (Steiner, Barbour and McCormick, 1984).

Soil compaction is a soil physical property that affects root development. Soil compaction alters the physical properties of the soil resulting in localized increases in soil bulk density which in turn affects soil air, water and temperature regimes (Rurark et al., 1982). Soil compaction can be caused by farm equipment or animals thereby destroying the soil's original structure (Broadfoot and Bonner, 1966). When sandy loam soil was compacted to a bulk density of 1.6, the root and shoot growth of planted cottonwood cuttings was retarded considerably. Root extension was below normal and hence shoot growth was

reduced. Broadfoot and Bonner (1966) concluded that soil compaction decreases the percentage of large pores with consequent decrease in aeration, moisture infiltration and movement of nutrients.

Soil compaction and cementation could also arise from "hardpans". Although several types of hardpans, differing in origin are known, this study was concerned only with ortsteins. Ortstein occurs when "all or part of the spodic horizon is at least weakly cemented when moist into a massive horizon that is present in more than half of each pedon" (U.S.D.A., 1975). Spodosol soil development is favored under vegetation cover with acid litter accumulation. Thus, spodosols occur under a wide range of trees including Tsuga, Picea, Pinus, Larix, Thuja, Populus, Quercus, and Betula (Buol, Hole and Mccracken, 1980).

According to Simonson (1968), in 1887 Muller was the first person to recognize the layer in which mobile substances had accumulated. Muller called these cemented or partially cemented B horizons: Ortsteins. Thus the term ortstein refers to the B horizon and is derived from two German words meaning "stone in place" (Winters and Simonson, 1951).

Simonson (1968) reported that ortsteins affect tree growth adversely and are troublesome when the soils are cultivated. Ortsteins occur in coarse textured soil material such as sands, coarse sands, gravels or mixture of these (Winters and Simonson, 1951). Generally the ortsteins in the northern U.S., as well as those in Europe, resemble a

conglomerate or concrete in general structure, with the coarse particles cemented together by iron oxides to form hardpan layers (Winters and Simonson, 1951). In addition to iron, ortsteins are also rich in aluminum and manganese and are not homogenous in chemical composition (Polskiy, 1961). In addition to the chemical heterogeneity, the morphology and color change with different degrees of waterlogging and with the length of time of excessive wetness (Oglenznev, 1968). Franzmeier and Whiteside (1963a, b) studied a 10,000-year-long chronosequence of spodosols in Michigan and concluded that between 3,000 and 8,000 years were required for the formation of a spodosol soil. Generally ortstein layers occur from 20 to 70 cm below the ground surface and their depth is not related to relief of the site (Muir, 1961).

MATERIALS AND METHODS

Ortstein layer

In the summer of 1984 intensive soil sampling was done to determine the distribution, depth and thickness of the ortstein layer in the two study areas. Soil borings one meter deep on transects 10 m apart were made using a soil augur. The first soil sample was taken 5 m inwards from the first tree between rows one and two. Thereafter, samples were taken 10 m apart for 150 m for a total of 15 samples per row. Subsequent samples were taken between every third row of trees. Each soil sample was checked for color, mottling and for presence or absence of a cemented ortstein

layer. When an ortstein layer was encountered, its depth from the surface and its thickness were noted. Following the above sampling procedure, a total of 180 and 120 sample holes was dug at the Mason and Manistee study sites, respectively (Figures 1 and 2). The minimum depth (the distance between the soil surface and the beginning of the ortstein layer) the maximum depth (the distance between the soil surface and the ortstein layer) and its thickness (the difference between the maximum and minimum depths) were compared within a study site and between the two study sites.

Soil nutrient levels

In the summer of 1986 soil samples were taken from both study sites for nutrient content determination. With the use of a soil augur, 10 holes at the Mason site and 7 holes at the Manistee site were dug one meter deep. Due to the size and shape of the study sites, a v-shaped sampling pattern was used at Mason (Figure 1) while samples were taken diagonally from the SE corner to the NW corner at the Manistee plantation (Figure 2). Soil horizons in a profile were differentiated by soil color or texture change as the plow layer (Ap), zone of eluviation (E) or zone of illuviation (B). The B horizon was further grouped as Bhs or as Bs based on the accumulation of organic matter (h) and sesquioxides of iron and aluminum (s). Each horizon depth, thickness and color were recorded. Color was for moist soil

conditions. Using Munsell soil color charts, gray or light gray horizons below the plow layer with hue of 7.5YR, 5YR or 2.5YR and a value of greater than or equal to four and a chroma of less than or equal to two were considered as E horizons. Horizons with 7.5YR or redder (5YR or 2.5YR) hue, with a value and chroma of <3/3 and >4/4 were grouped as Bhs and Bs horizons respectively. Soil samples from each horizon were tested for pH, nitrate nitrogen (NO3-N), extractable phosphorus (P), exchangeable cations: calcium potassium (K), magnesium (Mg); and exchangeable manganese (Mn), iron (Fe) and aluminum (Al). Soil pH was determined by the Glass Electrode pH Meter (Mclean, 1980). Nitrate nitrogen was determined using Quickchem method (Quickchem Systems, 1987), while the Bray P-1 test was used to determine extractable P level (Knudsen, 1980). Exchangeable Mn and Fe were extracted using 0.1N HCl (Whitney, 1980). Exchangeable K (Knudsen, Peterson and Pratt, 1982), exchangeable Ca and Mg (Lanyon and Heald, 1982) were determined using neutral 1N ammonium acetate (1N NH4OAc) extraction procedures. These nutrients and pH were determined by the Michigan State University Soil Testing Laboratory in East Lansing, Michigan. Exchangeable aluminum (Al +++) level was determined in the Forest Soil Laboratory, Michigan State University, using the DC-Argon plasma spectrophotometer (Barnhisel and Bertsch, 1982) (Appendix B).

Soil nutrient levels between study sites and within a study site were investigated. In addition, differences in nutrient levels between horizons were compared. Soil nutrient levels and disease ratings data from the two study sites were combined for comparison of soil nutrient with pest incidence ratings.

Methodology to assess relationship between soil characters and pest problems

Ortstein layer

Nine trees around each ortstein sampling point were identified to clone (Figure 1 and 2). Trees were grouped by the presence or absence of ortstein layer. Average tree height and percent mortality were compared between the two groups. The relationship between depth to the ortstein layer and tree height, canker incidence, and insect borer damage ratings were investigated.

Soil nutrient levels

Twenty trees around each soil sampling point for nutrient level tests were identified to clone (Figures 1 and 2). Possible correlations with soil nutrient levels and tree heights, canker incidence and, insect borer ratings were determined. Correlations between nutrient levels in the different horizons and disease ratings in a study site were determined. Using the Statistical Package for the Social Sciences (SPSS) (Niel et al., 1975), Scheffes multiple range test was used to test for significant differences in

nutrient level means between horizons in a profile or between profiles in a study site. All significant differences were indicated at the 0.05 level (Steel and Torrie, 1980).

Using the Number Cruncher Statistical System (NCSS) (Hintze, 1987) multivariate analyses were conducted by the stepwise regression method to determine how disease ratings were related to site factors and tree heights. "Best" multiple sets were selected on the basis that the partial correlation coefficients of the included variables were significant and the included variable substantially contributed to the explained variation.

RESULTS AND DISCUSSION

Ortstein layer

At the Mason study site 71 (39%) of the 180 borings and at the Manistee site 52 (43%) of the 120 borings showed an ortstein layer. The distribution of the ortstein layer was discontinuous in both sites. A one way analysis of variance for areas with ortstein showed that there were not significant differences in the minimum depth, maximum depth or thickness of the ortstein layer for the Manistee or Mason sites at P = 0.05. Comparison between the two sites showed that there was no significant difference in the beginning (minimum depth) of the ortstein layer (P > 0.05), while significant differences were found in the maximum depth of the ortstein layer (P < 0.05) and the ortstein layer

thickness (P < 0.05). The minimum, maximum and average depths of the ortstein layer are shown in Tables 9,10 and 11 for the Mason and Manistee sites and the comparison between the two sites respectively. The color of the ortstein layer, in a profile, varied from dark brown or reddish brown to pale yellow. Generally the darker ortstein layers were harder and more difficult to bore with a soil augur than the lighter colored layers. The pale yellow ortstein layer was more common at the Manistee site, where the water table was closer to the ground surface and soil mottling was observed, than at Mason. The conglomerate characteristic of an ortstein layer is shown in Figure 3.

Table 9. Summary of ortstein layer distribution and depth in cm at Mason plantation.

		Prof.	MIN.DEPTH		MAX.D	MAX.DEPTH		ORT.THICKNESS	
Trans	sect	with				Std. error		Std. error	
	 1	2	32.5	7.5	87.5	12.5	55.0	5.0	
	2	2	44.0	14.0	70.0	10.0	26.0	4.0	
	3	3	33.0	4.5	67.7	7.3	34.7	9.5	
	4	2	32.5	7.5	65.5	9.5	33.0	2.0	
	5	4	38.5	9.2	69.2	6.4	30.8	9.2	
	6	7	31.7	6.9	78.4	7.9	46.7	6.6	
	7	4	24.8	3.8	62.2	8.4	37.5	7.9	
	8	4	27.8	3.3	71.0	4.1	43.3	7.0	
	9	7	30.0	5.8	77.6	9.2	47.6	9.2	
	10	8	39.0	6.0	73.5	6.9	34.5	6.6	
	11	7	31.1	5.6	79.0	8.9	47.9	7.7	
	12	6	49.7	7.8	80.5	9.0	30.8	2.9	
	13	3	40.3	17.4	77.0	13.3	36.7	14.2	
	14	7	30.3	5.0	80.3	7.5	50.3	7.0	
	15	5	39.8	7.1	76.8	8.0	37.0	4.3	
r otal	180	71	34.8	1.9	75.4	2.2	40.6	2.1	

Table 10. Summary of ortstein layer distribution and depth in cm at Manistee plantation.

		MIN.	<u>DEPTH</u>	MAX.D	EPTH	ORT.T	HICKNESS
Transect					Std. error		
1	3	36.0	4.7	74.0	13.3	38.0	17.6
2	5	36.0	6.5	72.0	11.6	36.6	10.8
3	3	28.7	5.9	64.7	10.4	36.0	14.6
4					10.6		
5	7	31.4	6.0	77.7	7.5	46.3	10.7
6	2	25.0	2.0	43.0	2.0	18.0	0.0
7					13.8		
8	3	26.3	2.6	42.3	0.3	16.0	2.9
9	5	35.2	3.6	69.8	7.0	34.6	5.7
10					10.9		
11	4	28.8	5.2	70.8	11.0	42.8	7.9
12	2	20.5	2.5	38.0	4.0	17.5	6.5
13	2	52.5	22.5	74.5	25.5	22.0	3.0
14		31.0		65.0		34.0	
15		35.0			5.5		15.5
Total 120	52	33.1	1 . 8	67.0	3.0	33.9	2.8



Figure 3. The conglomeate characterstics of an ortstein layer, sample from the Mason plantation.

Table 11. Ortstein layer depth and thickness comparison between the two study sites.

		MIN.D	EPTH	MAX.DEI	PTH	ORT.THI	CKNESS
Site	Prof. with ort.	Mean	Std.	Mean	Std. error	Mean	Std. error
Maniste	e 52	33.1a	1.8	67.0a	3.0	33.8a	2.8
Mason	71	34.8a	1.9	75.3b	2.2	40.5 b	2.1
Total	123	34.0	1.3	71.8	1.8	37.7	1.7

^{*} means in each column followed by the same letter did not differ significantly at P = 0.05

Soil nutrient levels

The soil nutrient contents by study sites are shown in Tables 12 and 13 for the Mason and Manistee sites respectively. When soil sample means were combined over the study sites one way analysis of variance showed that there were significant differences in pH, P, K, Mg, Fe. Mn and Al levels between Manistee and Mason sites (Table 14).

No significant differences (P > 0.05) in nutrient levels between the ten sample profiles were observed at the Mason site (Table 15). At Manistee, however, N, Mg, and Al levels were significantly different in some of the seven sampling points (Table 16). Comparison of nutrient contents by horizon, however, showed that at Manistee only K was significantly different between the Ap horizon and the other horizons (Table 17), while in Mason pH, P, K, Ca, Mg, Fe,

Mn, and Al were found to be significantly different between the horizons (Table 18).

Except for N and Ca the two study sites were different in their soil nutrient levels. The Mason site had more P, K, and Mg than the Manistee site while the reverse was true for Fe, Mn and Al. More P, Fe and Al were found in the B horizons (where the ortstein layer was present) than in the Ap or E horizons at both study sites.

Table 12. Average soil nutrient contents (kg/ha) at Mason sampled from ten profiles (one m deep) with 32 horizons.

Nutrients	Min.	Max.	Mean	Std.error
рН	5.0	6.3	5.6	0.1
N	0.8	3.9	1.5	0.1
P	31.4	448.4	100.4	11.6
K	9.0	94.6	29.8	3.0
Ca	94.2	1699.3	587.5	53.0
Mg	17.9	299.3	108.7	8.8
Fe	4.5	142.4	33.8	4.0
Mn	2.2	15.7	3.6	0.4
Al	4.4	201.8	72.7	6.6

Table 13. Average soil nutrient contents (kg/ha) at Manistee sampled from seven profiles (one m deep) with 20 horizons.

Nutrients		Max.	Mean	Std.error
pН		7.4	5.4	0.1
N	0.8	3.7	1.7	0.2
P	9.0	74.0	22.4	2.4
K	9.0	33.6	17.3	1.6
Ca	188.3	1883.1	562.5	90.9
Mg	21.3	169.2	61.1	6.2
Fe	4.5	2470.5	389.1	101.6
Mn	2.2	20.2	4.5	1.0
Al	0.7	546.6	125.5	24.6

Table 14. Comparison of soil nutrient means (kg/ha) between the study sites.

	Ma	ason	Mani	stee	
Nutrient	Mean	Std. error	Mean	Std. error	Signi
рH	5.6	0.1	5.4	0.1	*
N	1.5	0.1	1.7	0.2	ns
P :	100.4	11.6	22.4	2.4	*
K	29.8	23.0	17.3	1.6	*
Ca !	587.5	153.0	562.5	90.9	ns
Mg :	108.7	8.8	61.1	6.2	*
Fe	33.8	4.0	389.1	101.6	*
Mn	3.6	0.4	4.5	1.0	*
Al	72.7	6.6	125.5	24.6	*

^{*} means significantly different at P=0.05 ns means not significantly different at P=0.05

Table 15. Variation in soil nutrient levels at the Mason plantation.

Pro				~~~~	-kg/ha				
		N	P	K	Ca	Mg	Fe	Mn	Al
1	5.5	1.0	87.6	29.9	487.6	140.7	24.3	4.5	62.1
2	5.7	1.2	170.6	23.9	645.1	91.4	47.8	3.0	45.0
3	5.6	2.1	68.9	51.6	548.1	67.3	74.5	3.4	69.2
4	5.6	2.2	122.7	40.1	384.2	125.5	37.6	4.5	83.7
5	5.3	2.2	66.7	33.1	498.5	60.5	47.1	5.0	86.0
6	5.5	2.1	77.6	22.6	438.5	68.2	30.0	3.6	108.7
7	5.9	1.3	171.2	25.5	637.2	137.0	29.1	2.8	37.3
8	5.7	1.0	79.2	27.7	660.5	126.1	18.5	3.9	71.2
9	5.6	1.1	65.6	21.3	731.7	161.7	17.4	2.8	90.2
10	5.5	1.1	72.6	27.7	613.7	112.1	11.8	2.8	85.3

^{*} no two profiles differ significantly at the P = 0.05 level by Scheffe's multiple range test.

Table 16. Variation in soil nutrient levels at the Manistee plantation.

Pro					ka/l	na			
				K	Ca	Mg	Fe		
1	4.8	3.0a	25.4			40.0a		2.6	302.3a
2	6.4	1.4b	18.7	22.1	942.7	124.4b	844.4	7.5	34.6a
3	5.1	2.0ab	18.7	17.1	440.9	62.8a	590.4	8.2	171.4a
4	5.2	1.2b	24.3	17.0	330.7	4 9.5a	47.6	2.8	120.8a
5	5.2	1.2b	10.1	25.2	377.7	64.4ab	186.1	2.2	63.4a
6	5.2	1.0b	21.3	12.0	251.9	21.3a	446.1	3.0	115.9a
7	6.4	1.0b	29.4	13.4	1249.8	68.4a	170.4	6.7	4.6b

^{*} soil nutrient means in each column followed by the same letter did not differ significantly at P = 0.05 level by Scheffe's multiple range test.

Table 17. Means of soil nutrients by horizon at the Manistee plantation.

						kg/ha	3			
	l Depth . in cm		N	P	ĸ	Ca	Mg	Fe	Mn	Al
Ap	0-22	5.4	1.8	16.8	26.9a	807.8	74.5	218.4	7.1	92.8
E	22-71	6.1	1.2	20.1	12.9ab	377.7	79.6	182.7	2.2	12.1
Bhs	22-58	5.3	2.1	21.5	16.1ab	613.8	59.4	780.1	5.0	233.7
Bs	52-100	5.6	1.3	30.3	11.9b	367.2	48.2	198.9	2.5	76.6

^{*} means in each column follwed by the same letter did not differ significantly using Scheffè's multiple range test at P = 0.05

Table 18. Means of soil nutrients by horizon at the Mason plantation.

Soi	l Depth					kg/l	na			
hor	. in cm	рН	N	Ь	K	Ca	Mg	Fe	Mn	Al
Ap	0-22	5.8a	1.4	118.5ab	62.3a	1144.9a	160.1a	31.2a	8.1a	23.3a
E	21-45	5.9a	1.4	71.1a	25.3b	694.1b	147.5a	11.2a	2.2b	37.9ac
Bhs	35-57	5.5ab	1.8	191.2b	30.5b	619.5b	138.0a	70.2b	2.2b	102.9b
Bs	49-100	5.3b	1.3	60.0a	15.1b	282.9c	62.5b	25.0a	2.4b	92.2bc

^{*} means in each column followed by the same letter did not differ significantly using Scheffè's multiple range test at P = 0.05

Relationship between soil characters and pest problems

Ortstein layer

Trees were grouped and compared by the presence or absence of an ortstein layer. A t-test of average height of trees between the two groups showed no significant differences at P = 0.05 at Manistee. The average height for trees in the presence of an ortstein layer was 3 m while it was 3.1 m in the absence of an ortstein layer (Figure 4). At Mason, however, there was a significant difference between the average height for trees growing in the absence of an ortstein layer (4.8 m) compared to 3.6 m for trees where an ortstein layer was present (Figure 5). mortality , however, was not different between trees growing in the absence or the presence of an ortstein layer at either site. Percent of dead trees was higher at Manistee (40% when ortstein present, 43% when ortstein absent) than at Mason (14% when ortstein present, 15% when ortstein absent).

Root development and penetration was severely restricted when the ortstein layer was present as shown in Figures 6 and 7.

The relationships between pest ratings and the effective soil depth (the soil depth to the cemented ortstein layer) are shown in Figures 8, 9 and 10 for the Mason plantation and in Figures 11,12 and 13 for the Manistee plantation. The effect of soil depth on the incidence of pests was probably due to its direct effect on tree heights. The direct

relationship between tree heights and pest incidence are shown in Figures 14, 15 and 16 for the Mason plantation and Figures 17,18 and 19 for the Manistee plantation.

Genrally the incidence of Septoria canker increased with increasing soil depth and tree height. Cytospora canker and poplar and willow borer incidences, however, tended to decrease or not change with increasing soil depth and tree height.

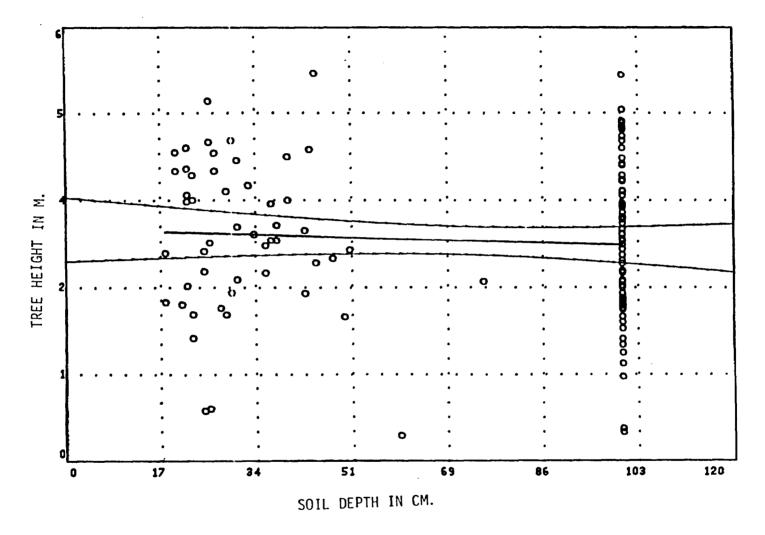
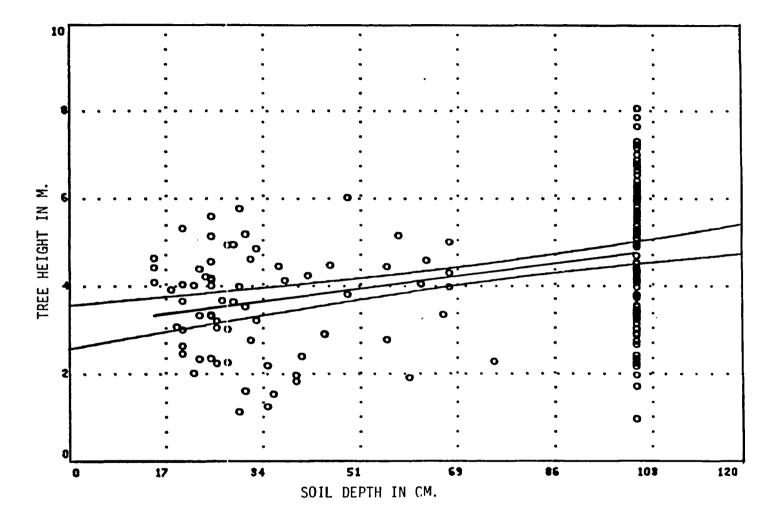


Figure 4. Relationship between soil depth to the ortstein layer and tree height at the Manistee plantation.



Figuer 5. Relationship between soil depth to the ortstein layer and tree height at the Mason plantation.



Figure 6. Root development and pattern at a place where the ortstein layer was absent at Mason. Note: photograph shows root upside down.



Figure 7. Root development and pattern at a place where the ortstein layer was present.
Note: photograph shows root upside down.

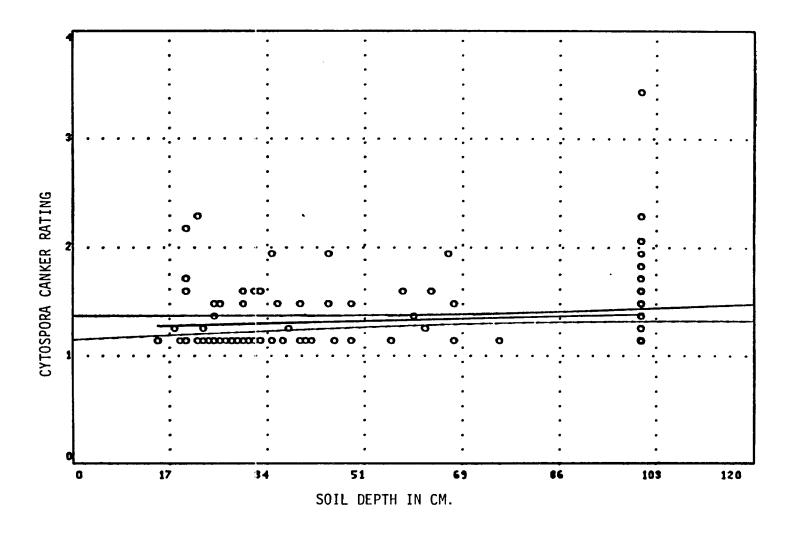


Figure 8. Relationship between soil depth to the ortstein layer and Cytospora canker rating at the Mason plantation.

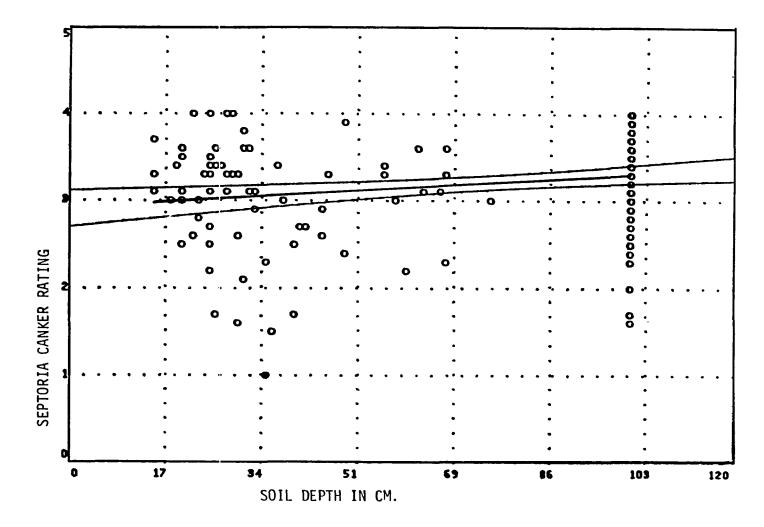


Figure 9. Relationship between soil depth to the ortstein layer and Septoria canker rating at the Mason plantation.

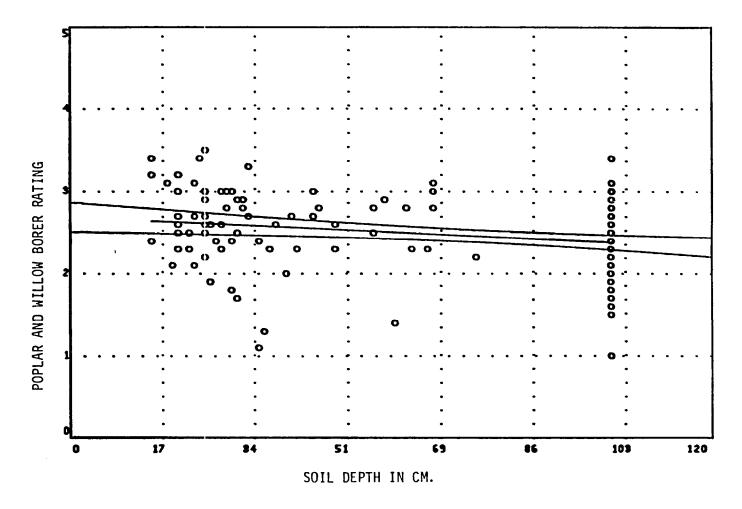


Figure 10. Relationship between soil depth to the ortstein layer and poplar and willow borer damage rating at the Mason plantation.

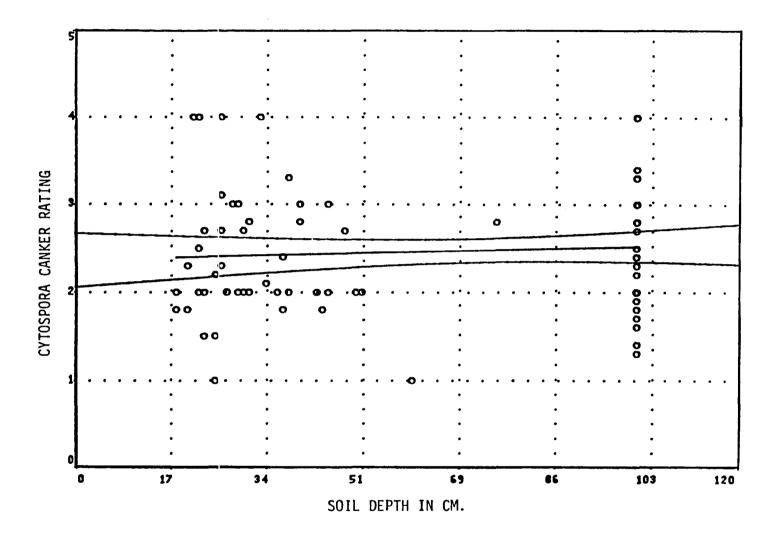


Figure 11. Relationship between soil depth to the ortstein layer and Cytospora canker rating at the Manistee plantation.

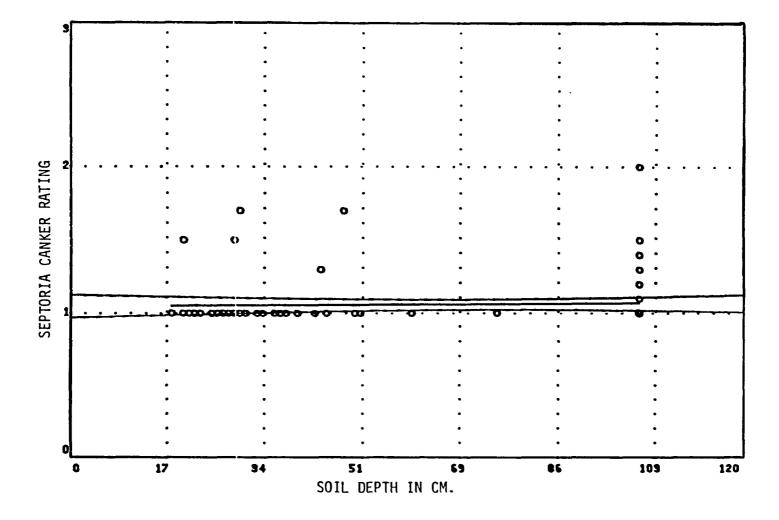


Figure 12. Relationship between soil depth to the ortstein layer and Septoria canker rating at the Manistee plantation.

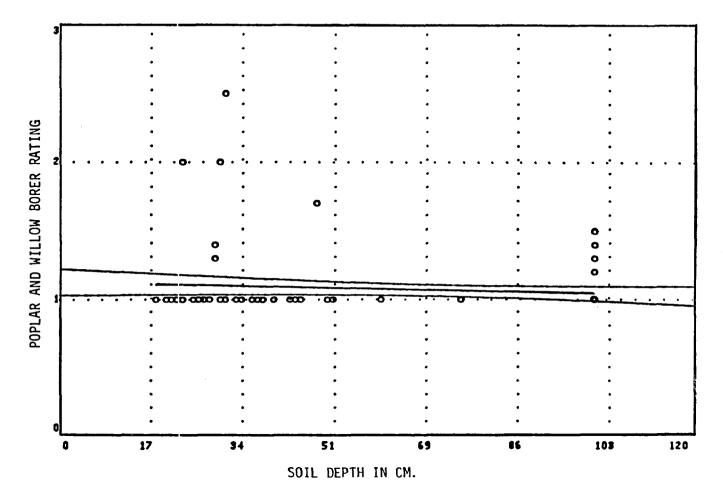


Figure 13. Relationship between soil depth to the ortstein layer and poplar and willow borer damage rating at the Manistee plantation.

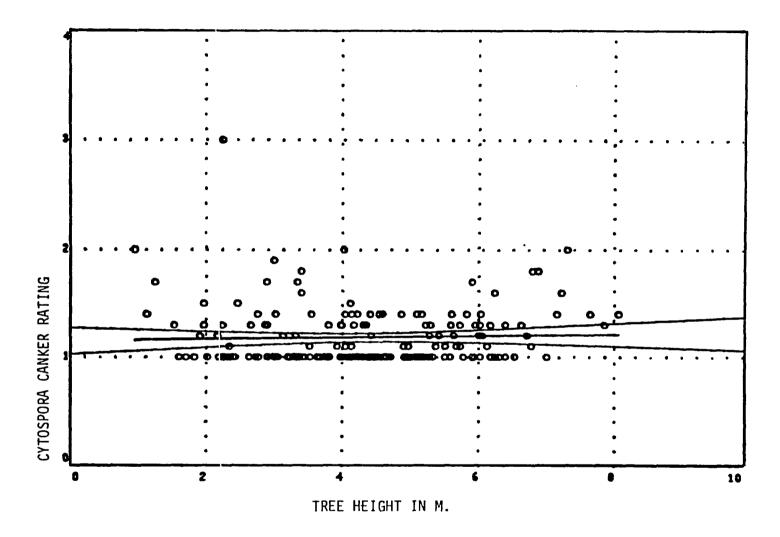


Figure 14. Relationship between tree height and Cytospora canker rating at the Mason plantation.

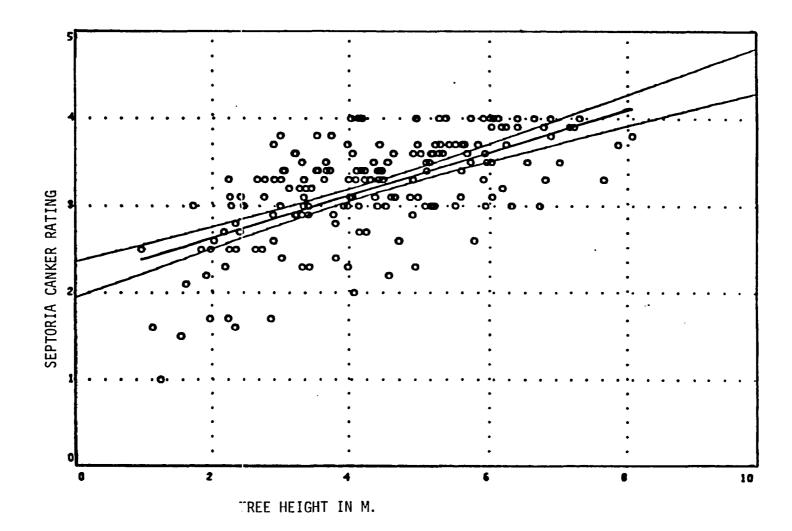


Figure 15. Relationship between tree height and Septoria canker rating at the Mason plantation.

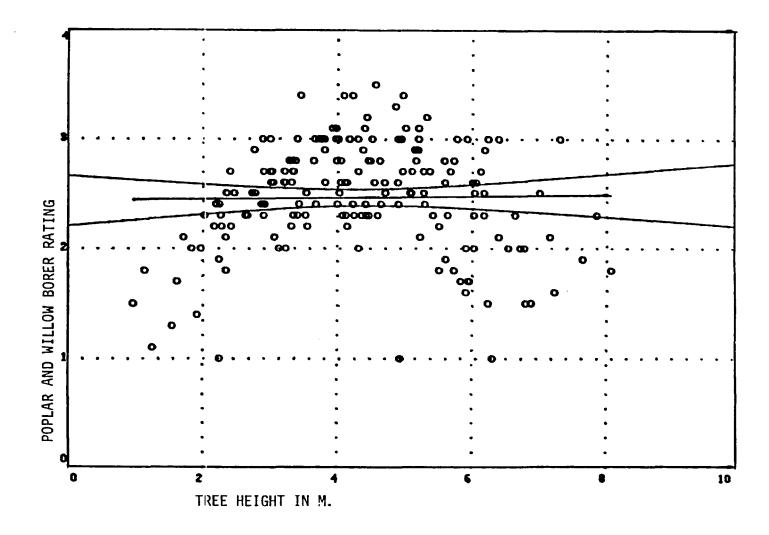


Figure 16. Relationship between tree height and poplar and willow borer damage rating at the Mason plantation.

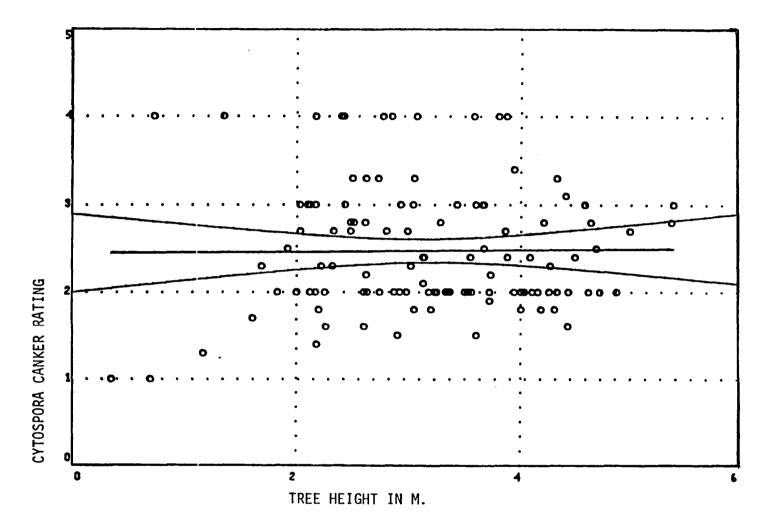


Figure 17. Relationship between tree height and Cytospora canker rating at the Manistee plantation.

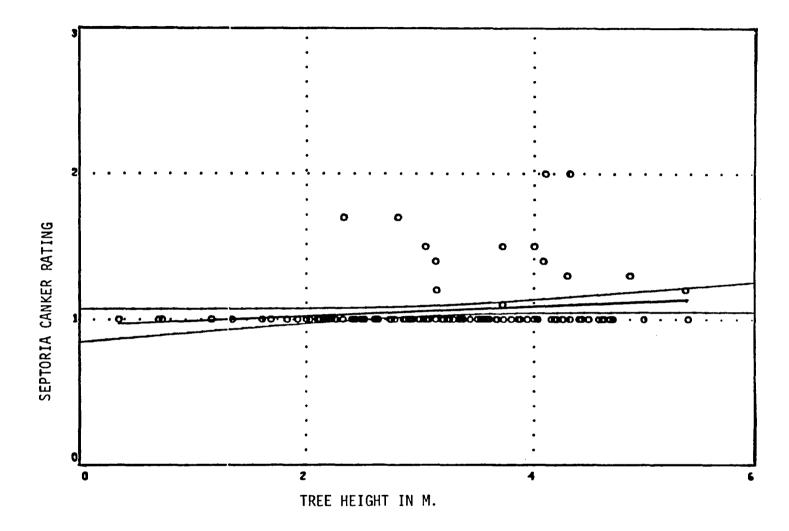


Figure 18. Relationship between tree height and Septoria canker rating at the Manistee plantation.

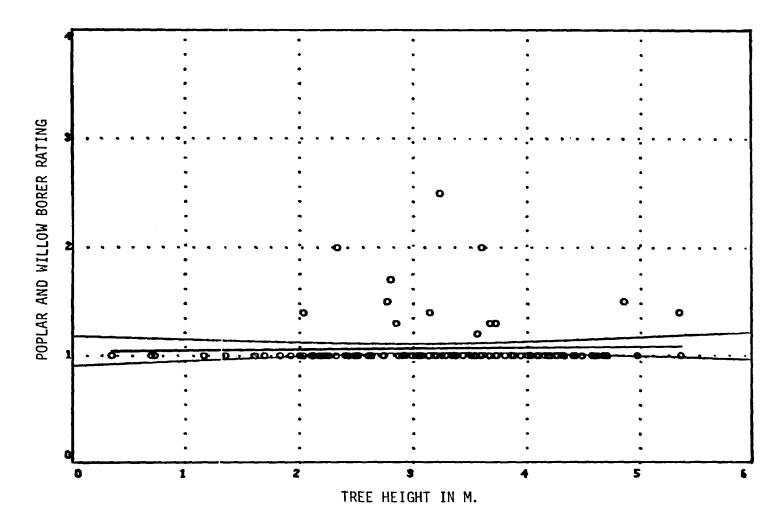


Figure 19. Relationship between tree height and poplar and willow borer damage rating at the Manistee plantation.

Soil nutrient levels

Pest ratings were correlated with soil nutrients levels at each horizon. Simple correlations between soil nutrient levels by horizon and pest ratings are presented for the Mason (Table 19) and Manistee (Table 20) plantations. In Table 21 soil nutrients that showed significant differences in their means when grouped by pest ratings are presented. In Table 21 the soil and pest ratings data were combined over the two study sites. Table 21 showed that Cytospora canker incidence increased as Fe and Al levels increased and K level decreased. Septoria canker incidence, however, increased with increasing levels of P, K and Mg and as Fe and Al levels decreased, while poplar and willow borer damage increased with increasing P and K levels.

In French prune orchards in California Cytospora canker occurance was associated with low level of K in the soil (Bertrand, English and Carlson, 1976). As Al level increased nutrient uptake of N, K, P, Ca and Mg by birch seedlings decreased (Goransson and Eldhuest, 1987) thereby causing an Al-induced nutrient deficiency (Godbold, Fritz and Huttermann, 1988). A similar process is presumed to happen at the two study sites. As Cytospora chrysosperma favors stressed trees, the high levels of Fe and Al and the low level of K lowered the tree's vigor and predisposed them to the fungus. Septoria musiva on the other hand preferably attacks vigorously growing tree's thus, the incidence of Septoria canker increased with increasing levels of P, K and

Mg (nutrients important for plant vigor and growth), and with decreasing levels of Fe and Al.

Table 19. Correlation coefficients between soil nutrient levels by horizon and disease ratings at Mason

CYTO	SPORA	CANKER	SEP	TORIA C	ANKER		BORER	
NE47	NE235	NE308	NE47	NE235	NE308	NE47	NE235	NE 308
Ap horiz								
PH -0.41 N -0.67* P -0.09 K -0.29 Ca -0.23 Mg -0.36 Fe 0.61* Mn 0.00	0.51 -0.32 -0.26 0.05 0.08	0.29 -0.28 -0.05 -0.28 0.34 0.23 0.35 0.13	-0.88* -0.98* -0.93* 0.54 0.60*	-0.36 -0.55 0.46 0.38 -0.19 -0.27	-0.41 -0.01 -0.51 0.23 0.14 -0.51	0.14	0.14 0.71* -0.04 0.14 -0.32 0.23 -0.31	-0.24 -0.01 0.37 0.35 -0.56 -0.50 -0.07 0.13 -0.03
E horizon								
pH N P K Ca Mg Fe Al		-0.49 -0.29 0.53 0.03 -0.34 0.02 -0.60 0.96*		0.66 -0.12 -0.64			-0.73 -0.36 -0.66 0.12 0.64 -0.26 -0.58	0.37 0.25 -0.48 -0.30 0.26 -0.38 0.22 -0.96*
Bhs horiz								
pH N P K Ca Mg Fe Al	0.21 -0.50 -0.29 0.57 0.16 0.65 -0.32	0.00 -0.36 -0.24 0.33 -0.09 0.64 -0.66		-0.56 0.60 0.02 -0.57 -0.67 -0.55 -0.28 0.60	0.32 0.38 0.54 -0.81* 0.34 -0.06 0.11 -0.17		0.15 0.24 0.65 -0.64 0.33 -0.26 0.36	0.45 0.12 0.62 -0.72* 0.41 0.49 -0.35
Bs horizon								
PH 0.56 N -0.52 P 0.01 K 0.26 Ca 0.30 Mg 0.12 Fe -0.93* Mn -0.16 Al -0.87*	-0.27 -0.14 -0.61* -0.14 0.01 0.14 0.09	0.05 -0.25 -0.12 0.05 0.18 0.17 -0.38 -0.18	0.92* -0.92* 0.76* -0.36 0.99* 0.95* -0.54 0.86* 0.23	-0.03 -0.29	0.11 -0.39 0.54 0.07 0.49 0.43 -0.14 0.72*	-0.58 0.53 0.54 -0.39 -0.38 -0.16 0.89* 0.00 0.78*	0.12 -0.05 -0.22 0.09 -0.00 0.08 -0.02 -0.06 0.01	-0.12 -0.11 0.14 -0.45 -0.58 -0.31 0.20 -0.23 -0.02

^{*} correlation coefficients significant at P = 0.05

TABLE 20. Correlation coefficients between soil nutrient levels by horizon and Cytospora canker incidence on NE 47 at Manistee.

	Ap horizon	Bhs horizon	Bs horizon
рН	-0.41	-0.71	-0.19
N	0.75*	0.64	0.74
P	0.27	0.76	-0.47
K	0.43	-0.15	-0.39
Ca	-0.35	-0.35	-0.30
Mg	-0.38	-0.46	-0.33
Fe	0.66	-0.50	-0.37
Mn	-0.40	0.02	-0.18
Al	0.21	0.77	-0.04

^{*} correlation coefficients significant at P = 0.05

TABLE 21. Means of soil nutrients grouped by pest ratings

				kg/h		
Pest	Rati	ng** P	K	Mg	Fe	Al
Poplar and	1	29.0a	30.0a			
willow borer	2	101.4b	61.1ab			
porer	3.	132.9b	65.5b			
Cytospora canker	1		63.5a		28.9a	22.5a
	2		31.2b		113.9ab	63.7ab
	3		25.2b		281.3b	134.9b
	4		33.6ab		448.4b	
Septoria	1	16.8a	26.9a	74.6a	218.4a	92.8a
canker	3	127.9b	66.1b	140.2b	37.6ab	22.6b
	4	30.7b	47.1b	239.3c	5.6b	26.3ab

^{*} means followed by the same letter within a column were not significantly different by Scheffe's procedure at P = 0.05

**	<u>Septoria</u> <u>and</u>	Poplar and
Rating	Cytospora Cankers	willow borer
1	Absence of canker	No apparent injury
2	Cankers on branches	Frass on the lower 1/3 of tree
3	Cankers on main stems	Frass on the lower 2/3 of tree
4	Main stem dieback, tree dead or dying	Frass on the lower 2/3 and the upper 1/3 of tree

Multivariate analyses for the three clones in the two study sites showed that soil nutrient levels and tree heights were the factors most related to the incidence of Cytospora and Septoria canker, and poplar and willow borer damage. The following were the variables in the regression equations that "best" predicted pest incidences at the two study sites:

Dependent_variables

Y₁= Cytospora canker on NE 235 at Mason

 Y_2 = Cytospora canker on NE 47 at Mason

 Y_3 = Cytospora canker on NE 47 at Manistee

 Y_A = Poplar and willow borer on NE 47 at Mason

 Y_5 = Septoria canker on NE 308 at Mason

 Y_6 = Septoria canker on NE 47 at Mason

Independent variables

 $X_1 = K$ content in $kg/\hbar a$

 $X_2 = Ca$ content in kg/ha

 $X_3 = Mn$ content in kg/ha

 $X_4 = P$ content in kg/ha

 $X_5 = Mg$ content in kg/ha

 $X_6 = Al$ content in kg/ha

 $X_7 = N$ content in kg/ha

 X_8 = Fe content in kg/ha

 X_Q = Height of trees in m

The equations for Cytospora canker were

 $Y_1 = 1.1088 - 0.0186X_1 + 0.0005X_2 + 0.0545X_3$

 $Y_2 = 3.1061-0.0328X_1-0.0023X_2+0.0171X_4-0.0434X_6$

 $Y_3 = 1.3248 + 0.0007X_2 - 0.0758X_3 - 0.0107X_5 - 0.8697X_7 + 0.1743X_9$

The coefficient of multiple determination (R^2), which is the ratio of the model sum of squares to the adjusted sum of squares, was the primary index of the equations goodness of fit to the raw data (Hintz, 1987). R^2 for equations Y_1 , Y_2 and Y_3 were 0.8012, 0.8162 and 0.7069 respectively.

The equation for poplar and willow borer with an R^2 value of 0.9061 was:

 $Y_A = 1.1046 + 0.0018X_2 + 0.0197X_3 - 0.0078X_6 - 0.1804X_8 - 0.457X_9$

The equations for Septoria canker were

 $Y_5 = 1.6625 - 0.0096X_1 + 0.00124X_2 + 0.0014X_4 - 0.0054X_5 + 0.00443X_6 - 0.0099X_8 + 0.235X_9$.

 $Y_6 = 2.1213 - 0.0135X_1 + 0.0041X_5 + 0.3687X_9$

 ${
m R}^2$ values for equations Y $_5$ and Y $_6$ were 0.7158 and 0.8371 respectively.

DISSCUSSION

The relationships between stand factors (clone, study site, tree vigor and plantation condition), soil factors (ortstein layer, physical and chemical properties) and major (Cytospora chrysosperma, Septoria musiva Cryptorhyncus lapthi) are summarized in Figure 20. lines show either a positive (+) or a negative (-) correlation within or between factors. Some of significant (P < 0.05) correlations include: at the Mason plantation there was positive correlation between Cytospora canker incidence and P, Mg, Mn and Al levels in the soil, while Septoria canker showed a negative correlation with K, P, Fe and Mn levels. Poplar and willow borer, however, was positively correlated with Mg level and negatively correlated with Al level in the soil. At the Manistee plantation Cytospora canker occurence was positively correlated with P, Fe, Mn and Al levels. As Figure 20 was made from the analysis of the correlations between site factors and pest incidences, it may indicate the possible relationships between the different factors, but it would not prove a cause and effect relationship. Furthermore, site factors other than those investigated may be important to the pest frequencies in the plantations.

The performance and productivity of a hybrid poplar plantation depends on the clones planted, site factors and the extent of pest damage. NE 47, which was planted at both study sites, grew faster at Mason than at Manistee,

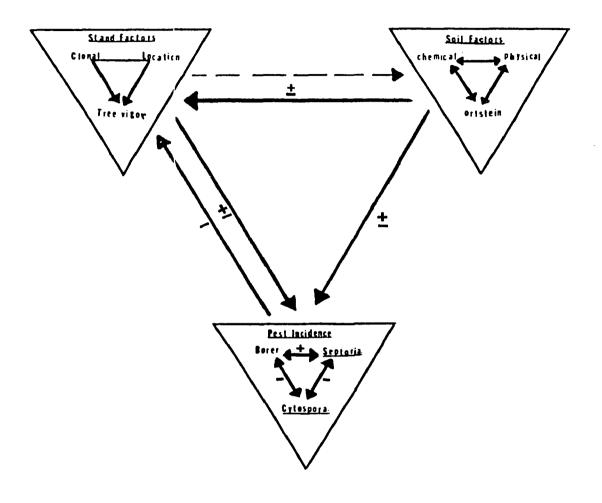


Figure 20. An interaction model between site factors and pests incidence on hybrid poplar clones at the Mason and Manistee plantations.

indicating inherent difference(s) between the two sites. The Mason site had more P, K, Mg and less Fe, Mn and Al than the Manistee site. Thus, the Manistee site with its high water table, poorly drained soil, and high levels of Fe and Al was more stressful to the trees than the Mason site, hence more Cytospora canker was found. A high level of Al in the soil may have been directly toxic to the roots or indirectly caused Al-induced deficiency of nutrients like Mg, K and Ca. The relatively slower growing trees at Manistee were thus predisposed to Cytospora canker, while Septoria canker and poplar and willow borer were more prevalent at the Mason plantation where the trees grew relatively faster and vigorously.

As the clones in the two study sites were not screened for insect or disease resistance, susceptible clones were planted and thus the trees were severely infected by pathogens or damaged by insects. The general conclusion of many studies was that the vulnerability to some canker pathogens increases in areas where soil moisture is low, particularly during stress periods (Bier, 1964). The presence of an ortstein layer reduced water percolation in early spring, thus causing waterlogging and physically prevented water absorbtion from a deeper level by the roots during the dry periods of the year, thus creating drought.

Therefore, before large areas of poplars are planted, their disease and insect susceptibility must be determined. To this effect Ostry (1981) suggested the following:

- incorporating disease resistance into tree screening and improvement programs.
- use of cultural practices which would not predispose the trees to pathogens (like deep plowing or subsoiling to break the ortstein layer).
- monitoring the plantations in order to detect pest problems early.
- determine critical level of infection and develop emergency measures to prevent large losses.

Generally, proper matching of clones to the site results in vigorous, fast growing plantations which are less suseptible to pests. However, fast growing trees could be more susceptible to aggressive pathogens like <u>S. musiva</u>. Usually the severity of pests and their impacts varies widely from clone to clone in poplars and depends mostly upon site (Barkley, 1983). Taking this into consideration the Ontario Ministry of Natural Resources (1983) advises: "to establish plantations by matching the most suitable clone to each particular site, plant several rather than one or two clones on each site and make clonal blocks in the plantations, each block being not more than five hectares." Thus, unlike the PCA large-scale mixed clone plantations, a geographic mosaic (Pinon, 1984) was a better alternative to pest management.

The genus <u>Populus</u> offers a diverse genetic make up. But by developing and planting only few clones, man has successfully limited the genetic base for disease resistance, adaptabilty to site and harsh environment. The

two plantations, investigated in this study, were even-aged with one to three clones planted intermixedly. Historically, pests have caused the failure of plantations which had a limited genetic base. Thus, these plantations were destined for failure due to pathogens, insects, environmental stresses or their combined interaction. Hence, once more, this study showed the importance of pests in plantation culture, and the risk of even-aged, narrow genetic based plantations.

The hybrid poplar plantations established in the past have a narrow genetic base, in the future broadenning their genetic base through selection, especially for disease resistance, from natural population might be essential. Planting hybrid poplar clones with a high degree of resistance to pests is undoubtedly a good management strategy.

In order to fully benefit from the fast growing habits of hybrid poplars and to minimize the loss of yield from pests; in the short term, an Integrated Pest Management (IPM) program with resistant clones as its cornerstone must be developed. The resistant clones should be obtained through local screening trials. In establishing and maintaining plantations silvicultural practices that would alleviate environmental stresses should be practiced. Before large-scale hybrid poplar plantations are established, more basic and applied researches should be conducted. Among others, these researches should include:

- -Studies on the heritability of resistnace to disease in poplars and the kind and number of genes involved.
- -Studies on the biochemical bases of pathogenicity of the major pathogens.
- -On regional and local bases, investigations on effects of the environment on the plant, on the pathogen and host-pathogen relationship, and particularly, the effect of soil chemical and physical properties on the host-pathogen relationship along with their effect on yield and growth habits of hybrid polars should be undertaken.
- -Since pest incidences vary by clone and location in hybrid poplars, widespread clone-site trials over a broad range of sites, locations and over the entire rotation period should be investigated.

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APPENDIX A

Procedure for estimating starch level in the roots

- 1. Dig root samples within 30 to 50 cm of the stem base. Take two root samples from horizontal roots that are approximately equal in diameter (one to two cm), as starch content is highly correlated to root diameter.
- 2. Cut root sample to 30 cms in length and put them in a polyethylene bag and keep in picnic ice box with ordinary ice to take back to the laboratory. Properly label samples.
- 3. If root samples are not to be analyzed immediately keep them frozen at -20° C.
- 4. Thaw root samples rapidly and prepare cross sections to approximately 50-100 μ thick with sliding microtome. Keep root cross sections moist at all times.
- 5. Put at least two cross sections on a glass slide and flood them with iodine (I_2KI) solution. Prepare Iodine solution by mixing 15g of Potassium iodine (KI) with 3g of crystalline iodine (I_2) in one litre of distilled water.
- 6. Blot excess solution and add fresh iodine solution.

 After five minutes blot the stain and rinse the root cross sections with distilled water.
- 7. Remove excess water, add glycerine and put a cover slip.
- 8. Record staining results on polaroid color film or 35 mm Extachrome (Kodak Co.) color slides. Pictures should be taken within 24 hours as stains fade through time.

9. Rate roots as high, medium, low or depleted in starch based on starch stain.

APPENDIX B

Procedure for exchangeable Aluminum extraction with 1N KCl.

- 1. Weigh 5g of soil into 125 ml erlmyer flask
- 2. Add 50 ml of 1 N KCl, stopper and shake for 30 min.
- Filter the supernatant solution through Whatman No. 42
 filter paper.
- 4. Collect filtrate to appropriate container for Al analyses.
- 5. Analyze for Al on the DC-Argon Plasma Spectrophotometer
- 6. Prepare standard of 5 ppm Al in 1N KCl extracting solution.

Prepare 5 ppm Al standard from 1000 ppm Al solution.

- a) Make 50 ppm from 1000 ppm Al solution
- b) Make 5 ppm from 50 ppm Al solution
- c) Use the 5 ppm Al as the standard