## SOFT-ROT CAPABILITIES OF THE MAJOR MICROFUNGI, ISOLATED FROM DOUGLAS-FIR POLES IN THE NORTHEAST<sup>1,2</sup>

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#### ABSTRACT

Four hundred seventeen fungi were isolated from 144 of the 163 Douglas-fir poles (ages 7 to 17 years and treatments CCA, penta and oil, or Cellon<sup>®</sup>) sampled from transmission lines or storage piles in New York and Pennsylvania. Microfungi predominated and comprised nearly 85% of all isolates. They were isolated primarily from treated zones and were most abundant in older CCA-treated poles in transmission lines. *Antrodia carbonica* and *Postia placenta* were the principal basid-iomycete decayers and isolated primarily from untreated zones in CCA-treated poles. A limited number of white-rot fungi were isolated from the treated and untreated zones of several poles.

Seven of the 12 principal microfungi were established to have soft-rot capabilities. Soft rot was detected anatomically in 23 of the 144 poles in transmission lines. In most cases it was superficial and limited to several outer annual rings; however, it was severe in older CCA-treated poles and involved all of the treated zone and extended several centimeters radially into the untreated zone. Also, soft rot was detected anatomically and soft-rot fungi culturally, in 8 of 12 13-year-old CCA-treated poles that had been fumigated with Vapam 5 or 6 years previously. None was detected in the fumigated poles.

These data suggest that soft-rot fungi play an important role in decay development in the treated groundline zone of utility poles and should be considered in decay detection programs (culturally) and decisions on the timing of remedial treatments.

Keywords: Decay, soft rot, microfungi, utility poles, Douglas-fir, CCA, penta and oil, basidiomycetes.

#### INTRODUCTION

Information on the identities and roles of the major fungi associated with aging treated utility poles in service and eventual decay development has several current

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and potential uses. It detects early decay, provides estimates of pole service lives, and judgments on decay origins and development rates. It may lead eventually to improved pole treatment, handling and maintenance as the vulnerable points and fungal protagonists of the pole protective system are better understood.

Previous studies have determined the major basidiomycete decayers in Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco] poles in the United States and Canada (Eslyn 1970; Zabel et al. 1980) and both the basidiomycete decayers and microfungi in southern pine (*Pinus* spp.) poles, in service in the eastern United States (Zabel et al. 1985). The latter study revealed that soft-rot damage was common in older creosote-treated pine poles and that several species of microfungi, isolated frequently from the older poles, were capable of soft rot.

This information, along with the known importance of soft-rot fungi in poles elsewhere (Henningsson et al. 1975; Greaves 1977; Levy 1978; Leightley 1980), and in other wood products (Duncan 1960; Nilsson 1973; Morrell and Smith 1988) raised the questions of the identities and soft-rot potential of the major microfungi in treated Douglas-fir poles in New York.

The objectives of this study were: a) to isolate, identify, and determine the softrot capabilities of the principal microfungi found in the groundline zone; b) to anatomically study wood from the groundline zone and to determine soft-rot frequency, location, and severity; and c) to determine any relationships among the species, and prevalence of the microfungi isolated, with preservative type, pole age, and radial position of the pole in the groundline zone.

## MATERIALS AND METHODS

## Pole selections

Douglas-fir transmission poles representing three preservative treatments and age classes (Table 1) were solicited from six utilities in New York. Availability or reasonable access limited the poles sought in some pole age-treatment groups. Small additional samples of storage and fumigated poles were added in the second year of the study. The poles sampled in the study totalled 163.

## Pole sampling procedures

Pole sampling was done in the summer and fall of 1984 and 1985. Pole brand or utility records<sup>3</sup> provided information on the preservative treatments, treaters, class, and pole age in service or since treatment, and the nature and timing of any remedial treatments.

A hole about 30 cm (12 in.) deep was dug adjacent to the deepest check in the groundline zone. The pole surface exposed was scraped clean and probed with a sharp instrument for soft zones. The depth and extent of surface-softened wood were recorded and its appearance described. A small surface sample (approximately 1 cm  $\times$  1 cm and 1–2 mm in depth) was removed for anatomical study. A core was taken aseptically about 15 cm below ground (adjacent to the surface sample site) with an increment borer and placed in a sterile tube for later isolations

<sup>&</sup>lt;sup>3</sup> According to the utilities, the poles purchased met ANSI (1979) specifications and AWPA preservative and treatment standards in effect at that period.

		Preservative treatment and post-treatment ages (years)								
	Penta	and oil	Celi	lon*	C	CA	Totals			
Pole use or remedial treatment	0-10	11-20	0-10	11-20	0-10	11-20	0-20			
Transmission	22	40	_	27		35	124			
Transmission (fumigated)	_	8	_	-	-	12	20			
Storage	9		6	-	4	_	19			
Totals	31	48	6	27	4	47	163			

 TABLE 1. The distribution of the Douglas-fir transmission poles sampled in the several age, treatment and use groups.

and microscopic study in the laboratory. The sample bore holes were then flooded with a preservative, fitted tightly with a treated dowel, and the soil replaced. In the fall of 1985, nine CCA-treated poles were available for dissection studies. Samples were taken from radial slabs sawed from the groundline zone.

## Anatomical studies

The increment cores were studied with a stereomicroscope and features such as depth of preservative penetration, texture, color, and decay condition noted on full-scale core sketches.

Microscopic sections for the detection of soft rot were cut from radial and/or transverse planes of the surface chips or from the outer-treated zone of the increment cores or radial slabs. In those few cores or slabs where soft rot was detected throughout the radial dimension from the surface section, additional sections were prepared at 1-cm increments. In such cases of extensive soft rot, the 1-cm increments were judged from the original macroscopic sketches of each core or slab. Radial depths of soft-rot attack in the surface samples were measured microscopically using a calibrated eyepiece micrometer. Permanent slides were prepared after dehydration in an alcohol, alcohol-xylene series and mounted in Permount®. When soft rot was detected, cross sections were prepared from wood in the same position to obtain quantitative estimates of the intensity of the soft-rot attack. The radial and cross sections were studied microscopically to determine the types, locations, and degree of soft-rot damage in the cell walls (Wilcox 1964). In some cases, fiber suspensions were used also to detect or confirm cavities or cell-wall erosion in cases when the wood was difficult to section or contained copious amounts of residual preservative. Small slivers of wood were soaked in a mixture containing equal volumes of glacial acetic acid and 30% hydrogen peroxide and heated in a warm bath until fibers separated. The fibrous mat was rinsed several times in distilled water and then a small sample of fibers (50-100) was mounted on slides using a fixative (Meyer et al. 1988).

## Isolations and identifications

Isolations were made from three radial positions on the core (1. outer-treated; 2. inner-treated; and 3. untreated). The media used were 3.0% malt extract agar (general medium for microfungi) and a 1.25% malt extract agar containing 0.04% benomyl (selective medium for basidiomycetes). Tetracycline was added to all media (50 mg/liter) to reduce bacterial growth (Wang et al. 1989). Twelve chips were cut per core position and placed equidistant, three to a plate (two plates per

medium), so a portion of the chip penetrated the medium (12 plates or 36 chips per pole).

The plates were incubated at 28 C and reviewed at weekly intervals for up to 6 weeks for fungus development and subsequent subculturing. Reference slides were prepared for each isolate, which was then purified by streaking and/or re-isolation from culture margins. The isolates were maintained in malt extract agar (MEA) tube cultures for later study and identification.

Fungal identifications were made by microscopic study, comparison with known cultures, special cultural studies, and in a few cases by sending representative isolates to specialists for confirmations or identifications.

## Soft rot tests

Soft rot tests were conducted on 12 microfungi, selected on the basis of prevalence or close taxonomic relationships to important soft-rot fungi (Table 6). The test procedure used was a modification of the Nilsson method (1973).

Douglas-fir sapwood blocks,  $2 \times 2 \times 1.2$  cm (longitudinal plane) were labelled, oven-dried at 102 C for 24 hours to a constant weight (odw), and weighed to the nearest mg. The blocks were immersed in distilled water and a vacuum applied until a moisture content of approximately 100% was achieved. Two moistureadjusted blocks were placed side by side with a cross section surface downward in a 16-oz square bottle containing 10 g of Vermiculite. The Vermiculite had been sifted on a 5-mm mesh screen and the coarse particle fraction used. An additional 10 g of Vermiculite was added to cover the blocks. Fifty ml of a mineral-nutrient solution was added to each chamber (NH<sub>4</sub>NO<sub>3</sub>, 6 g; K<sub>2</sub>HPO<sub>4</sub>, 4 g; KH<sub>2</sub>PO<sub>4</sub>, 5 g; MgSO<sub>4</sub>· 7H<sub>2</sub>O, 4 g; glucose, 2.5 g; and 0.1 mg thiamine-HCl per liter of distilled water). The decay chambers were sterilized for 30 minutes at 121 C, allowed to cool for 24 hours, and resterilized at 121 C for 15 minutes.

After cooling for 48 hours, the chambers were inoculated with spore and/or mycelial suspensions prepared from each test fungus. The inoculum was prepared by aseptically placing a 3-mm-diameter plug of mycelium cut from the margin of an actively growing 7-day-old culture into 10 ml of a 2.5% malt extract solution, incubating without agitation for 7 days at 28 C, and dispersing it with a tube stirrer just prior to introduction into the decay chambers.

The inoculated decay chambers were incubated for 6 months at 24 C or 32 C. Replicates of 10 blocks per test fungus per temperature group were used. Nine blocks were used to determine weight losses caused by a test fungus. The tenth block was used for anatomical studies of the fungus colonizing the block.

Noninoculated control blocks were used to determine any effects of the decay chamber exposure on odw's. Reference blocks were removed from the noninoculated chambers at monthly intervals and used to monitor block moisture levels.

At the end of the incubation period, the purity of the original inoculum in each decay chamber was verified by plating several Vermiculite granules on MEA and confirming the original isolate.

The test blocks were then removed, gently brushed clean, the wet weights measured, and the macroscopic appearances described. The odw's were obtained for 9 of the blocks per temperature-fungus group as described above. Weight losses were measured as the difference between the original and post-decay odw's and expressed as a percent of the original odw. The soft-rot capability of a test fungus was judged as positive when weight losses in the blocks exceeded 2% (the sapwood water-soluble extractive content) and/or the longitudinal bore holes or significant erosion were observed microscopically in the S2 zone of the cell walls of the anatomical study blocks.

The tenth block was fixed in a formalin-aceto-alcohol (FAA) solution and sectioned for anatomical studies. Permanent microscopic slides of radial and cross section surfaces were prepared as described above. They were studied microscopically also to determine the type, location, and extent of cell-wall damage.

Data handling. — The general data collected for each pole and the information on wood condition and associated fungi for the various radial positions in the groundline zone were coded and assembled for computer compilation. The data set was sorted for the effects of pole treatment and pole age on the types and frequency of the fungi isolated and the incidence and locations of basidiomycete and soft-rot decay in the poles.

## RESULTS

## Identities and groupings of the isolates

Four hundred seventeen isolates representing an estimated 70 species were obtained from the groundline zone of 144 of the 163 Douglas-fir poles studied. The isolates were subdivided by microscopic study into basidiomycete (66) and microfungi (351) groups,<sup>4</sup> and sorted by radial core position of isolations. The fungi are listed in decreasing prevalence, as currently identified to species, genus, taxon, or group in Table 2. Cultural and microscopic descriptions of these species and other common pole-inhabiting fungi have been reported elsewhere (Wang and Zabel 1990).

Antrodia (Poria) carbonica and Postia (Poria) placenta were the principal basidiomycete decayers and were obtained primarily from untreated heartwood zones and to a lesser extent the inner-treated zones in the poles. They were associated generally with a brown cubical rot in the untreated pole zones. The six additional species of basidiomycetes isolated were white-rot fungi (Table 2) and obtained from both treated and untreated zones.

Pachnocybe ferruginea traditionally classified as a microfungus but recently placed in the basidiomycetes (Kropp and Cordon 1986) was also isolated. It is a phenol oxidase producer so could be grouped with the other white-rot fungi. It was found most commonly in the untreated-core zone and was not associated with wood decay.

Microfungi were the principal fungi isolated from the poles and were obtained most commonly from the treated zones. The four microfungi isolated most frequently were a *Hyalorhinocladiella* sp., *Phialemonium dimorphosporum*, *Scytalidium lignicola*, and *Phialophora mutabilis*. All with the exception of the *Hyalorhinocladiella* sp. produced the soft-rot type of decay. Seventeen species of microfungi, some still unidentified as to species, were isolated four times or more from the poles and represented nearly half of the isolations. Four species of

<sup>&</sup>lt;sup>4</sup> Microfungi is a general term used for fungi with microscopic fruiting bodies. Many microfungi are in the Deuteromycetes (Fungi Imperfecti). Macrofungi such as the wood-destroying basidiomycetes have large fruiting bodies visible to the unaided eye.

	Number of isolations radial core positions						
Genus, species, taxon, or other taxonomic group	Outer-treated	Inner-treated	Untreated	Totals			
Basidiomycete de	cayers						
Antrodia carbonica (Overh.) Ryv. & Gilbn.	1	5	22	28			
Postia placenta (Fr.) M. Lars. & Lomb.		4	8	12			
Pachnocybe ferruginea (Sow.:Fr.) Berk.	1	2	5	8			
Bjerkandera adusta (Willd.:Fr.) Karst.			2	2			
Misc. basidiomycetes <sup>a</sup>	1	2	2	5			
Unknown basidiomycetes	2	3	6	11			
Microfungi							
Penicillium spp.	22	11	11	44			
Hyalorhinocladiella sp.	18	12	5	35			
*Phialemonium dimorphosporum W. Gams	14	10	6	30			
Scytalidium lignicola Pesante	14	8	0 7	27			
Phialophora mutabilis (Beyma) Schol-Schwarz	7	5	6	18			
Exophiala sp.	8	3	3	14			
Dematiaceous unknowns	6	4	1	11			
Phialophora sp. M	3	3	4	10			
Trichoderma spp.	2	3	4	9			
Alternaria alternata (Fr.) Keissler	6	1	4	8			
Scytalidium circinatum Sigler & Wang	3	2	3	8			
Phialophora sp.	4	2	1	7			
Taxon 93	4	3	2	7			
	4	1	2	7			
*Epicoccum nigrum Link	1	4	2	7			
Cladosporium herbarium (Pers.) Link	2	4	4	, 7			
Cladosporium sphaerospermum Penz.		2	-	6			
Moniliaceous unknowns	1	_	3 2	6			
Hormoconis resinae (Lindau) v. Arx & de Vries	3	1 2	4	6			
Paecilomyces variotii Bainier	3	2	4	6			
Zygomycetes	3 2	2	2	5			
*Phialophora malorum (Kidd & Beaum.) McColloch		2	1	4			
*Phialophora fastigiata (Lagerb. & Melin) Conant	1 2	1	1	4			
Imperfect unknowns	2	1	3	4			
Penicillium diversum Raper & Fennell		1	2	3			
Bispora betulina (Cda.) Hughes	1	1	2	3			
Phoma fimeti Brun.	1 2	1	1	3			
Rhinocladiella atrovirens Nannf.	_	1	-	3			
Talaromyces flavus (Klöcker) Stolk & Samson	1	1	1				
Phialemonium sp. A.	2	12	1	3			
Misc. Microfungi <sup>b</sup>	21	12	13	46			
Totals	158	118	141	417			

TABLE 2. The identities and numbers of the fungi isolated from the groundline zones of 144 of the 163 penta- or CCA-treated Douglas-fir transmission poles sampled in New York and Pennsylvania.

\* Basidiomycetes isolated one time were: Perenniporia tennuis (Schw.) Ryv.; Diplomitoporus lindbladii (Berk.) Gilbn. & Ryv.; Trametes

<sup>&</sup>lt;sup>a</sup> Basidiomycetes isolated one time were: Perenniporia tennuis (Schw.) Ryv.; Diplomitoporus lindbladii (Berk.) Gilbn. & Ryv.; Trametes versicolor (L.:Fr.) Pilat; Ptychogaster rubescens Boud.; and Irpex lacteus (Fr.:Fr.) Fr.
<sup>b</sup> Microfungi isolated two times or less were: Acremonium sp. A.; Acrogenospora sphaerocephala (Berk. & Br.) M. B. Ellis; Alternaria sp.; Anthrinium arundinis (Corda) Dyko & Sutton; Aspergillus spp.; Aureobasidium pullulans (de Bary) Arnaud; black yeast; Cephaloascus albidus Kurtzman; Chaetonnium funicola Cooke; Cladosporium spp.; Coelomycete; Curvularia sp.; Cylindrocarpon sp.; Exophiala sp.; Fusarium solani (Mart.) Sacc.; Fusarium oxysporum Schlect; Gliocladium sp.; Gliocladium roseum Bainier; Hormonema dematioides Lagerb. & Melin; Oidiodendron griseum Robak; Paecilomyces lilacinus (Thom) Samson; Phialocephala fusca Kendrick; Phialophora richardsiae (Nannf.) Conant; Phorma sp.; Scopulariopsis sp.; Stachybotrys elegans (Pidopl.) W. Gams; Sterile isolate; Talaromyces trachyspermus (Shear) Stolk & Samson; Taxon Phialemonium/Acremonium; Phialemonium sp.; \*Taxon 121; Taxon 146 (sterile); Trichoderma honigii Oud.; Trichoderma polysporum (Link) Rifai.

*Phialophora* were in this group. The microfungi that were isolated four or more times from the poles were considered potentially important and selected for further study. They are termed principal fungi hereafter in the data analysis.

The remaining microfungi were a large diverse group. Many were isolated only one or two times. Others in some groups such as some *Penicillium* species and the dematiaceous unknowns represent fungi of taxonomic complexity and are still unknown.

### Isolate groupings by pole status and preservative treatment

The principal isolates were grouped by preservative treatments and core positions within the three pole use groups (non-fumigated transmission poles in service, fumigated transmission poles in service, and stored poles) for any inferences on invasion routes and related preservative tolerances (Tables 3–5).

Non-fumigated transmission poles.—Antrodia carbonica and Postia placenta were isolated only from CCA-treated poles and primarily from the untreated zone. This supports prior speculations that these fungi enter the poles via deep checks during storage or seasoning and survive the preservative treatment (Zabel et al. 1982). Two white-rot fungi (other basidiomycetes) were obtained also from the untreated zone of the CCA-treated poles. However, Trametes versicolor, Irpex lacteus, and Diplomitoporus lindbladii were obtained only from treated zones in the older penta-treated poles, suggesting tolerance and entrance through the treated zone. In the microfungi, a frequently isolated species, Hyalorhinocladiella sp., was associated exclusively with the penta and oil treatment and primarily the treated zones. This suggests a tolerance to pentachlorophenol and association with the oil carrier since it was absent in the Cellon<sup>®</sup>-treated poles. A soft-rot fungus, Phialemonium dimorphosporum, was also obtained exclusively from the treated zone of the penta- and oil-treated poles. Scytalidium lignicola, a soft-rot fungus, was also obtained primarily from the treated zones of the penta-treated poles. In many poles, these two fungi were found primarily in the treated zones, suggesting some tolerance and invasion from the surface after penta depletion.

Several of the other soft-rot fungi (*Phialophora mutabilis, Alternaria alternata, Phialophora* sp. M and *Epicoccum nigrum*) were isolated only from CCA-treated poles, and primarily from treated zones suggesting some preservative tolerance or heartwood intolerance and again an outside-in invasion pattern in the ground-line zone. Also many of the minor fungi listed in Table 2, which accounted for much of the diversity and taxonomic complexity of the pole mycobiota, were obtained from the treated zones of the older CCA-treated poles.

Fumigated transmission poles. —A comparison of the fumigated penta- and oiltreated poles in Table 4 with their similar non-fumigated poles in Table 3 suggests that fumigation with Vapam was highly effective and no reinvasion of the poles occurred in the groundline zone for up to six years. A similar comparison of the fumigant-treated CCA poles in Table 4 with their reference poles in Table 3 suggests that all basidiomycete decayers were killed and have not reappeared for up to five years. However, it is important to note that substantial numbers of microfungi are now present in the fumigated poles and that eight are soft-rot fungi, including *Phialemonium dimorphosporum* and *Scytalidium lignicola*, the most prevalent soft rotters in the study. Whether these fungi represent reinvasion of the poles, fumigant treatment survivors, or a combination is unknown. Their

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				N	lumber of isolatio	ns <sup>b</sup>			
		Penta and oil			Cellon®			CCA	
Genus, species, or taxon of the principal fungia	Outer-treated	Inner-treated	Untreated	Outer-treated	Inner-treated	Untreated	Outer-treated	Inner-treated	Untreated
			Basidio	omycetes					
Antrodia carbonica							1 (3)	4 (11)	19 (54)
Postia placenta								2 (6)	7 (20)
Pachnocybe ferruginea		1 (2)	1 (2)	1 (2)	1 (2)	1 (2)			3 (9)
			Micr	ofungi					
Hyalorhinocladiella sp.	18 (29)	12 (19)	5 (8)						
*Phialemonium dimorphosporum	8 (13)	3 (5)	2 (3)						
*Scytalidium lignicola	11 (18)	1 (2)		1 (4)	6 (22)	5 (18)		1 (3)	
*Phialophora mutabilis							6 (17)	4 (11)	6 (17)
Exophiala sp.	1 (2)		1 (2)	1 (4)			6 (17)	3 (9)	2 (6)
*Alternaria alternata							4 (11)	1 (3)	1 (3)
*Phialophora sp. (Taxon M)							3 (9)	2 (6)	1 (3)
Taxon 93				1 (4)	1 (4)	1 (4)	1 (3)	2 (6)	1 (3)
*Epicoccum nigrum							3 (9)	1 (3)	1 (3)
Cladosporium herbarium							1 (3)	4 (11)	2 (6)
Cladosporium sphaerospermum							2 (6)	1 (3)	4 (11)
Hormoconis resinae						1 (4)	2 (6)	1 (3)	1 (3)
Paecilomyces variotii			2 (3)		1 (4)	1 (4)		1 (3)	
*Phialophora malorum									
*Phialophora fastigiata								1 (3)	1 (3)
Penicillium diversum							1 (3)	3 (9)	

TABLE 3. The numbers of the principal fungi isolated from three core positions in the groundline zone of 124 Douglas-fir transmission poles with three preservative treatments.

\* The principal fungi were defined as the fungi which appeared four times or more in the isolations from the 163 poles listed in Table 2. <sup>b</sup> Pole treatment sample sizes were: penta and oil, 62; Cellon \*, 27; and CCA, 35. The number of isolations was expressed as a percent of the sample size and placed in parentheses to facilitate comparisons across preservatives.

\* Fungi determined in this study to cause soft rot in laboratory decay tests.

	Number of isolates <sup>a</sup>								
	F	Penta and oi	l <sub>p</sub>		CCA <sup>b</sup>				
Genus, species, taxon or groups of fungi isolated	Outer- treated	Inner- treated	Un- treated	Outer- treated	Inner- treated	Untreated			
*Phialemonium dimorphosporum	0	0	0	5 (42)	7 (58)	4 (33)			
Penicillium spp.	0	0	0	6 (50)	2 (17)	1 (8)			
Trichoderma spp.	0	0	0	1 (8)	2 (17)	2 (17)			
Dematiaceous unknown	0	0	0	1 (8)	1 (8)	1 (8)			
Phialophora spp.	0	0	0	1 (8)	2 (17)	3 (25)			
Phoma fimeti	0	0	0	1 (8)	1 (8)	1 (8)			
Talaromyces flavus	0	0	0	1 (8)	2 (17)				
Acremonium sp. A.	0	0	0		1 (8)	1 (8)			
*Phialophora fastigiata	0	0	0	1 (8)	1 (8)				
*Phialophora malorum	0	0	0	1 (8)		1 (8)			
Phialophora mutabilis	0	0	0	1 (8)	1 (8)				
*Scytalidium lignicola	0	0	0			2 (17)			
*Alternaria alternata	0	0	0	1 (8)					
Black yeast	0	0	0			1 (8)			
*Epicoccum nigrum	0	0	0			1 (8)			
Paecilomyces lilacinum	0	0	0	1 (8)					
Penicillium sp. A	0	0	0	1 (8)					
Penicillium sp. B	0	0	0	1 (8)					
*Phialophora sp. Taxon M	0	0	0	1 (8)					
Scopulariopsis sp.	0	0	0	1 (8)					
Trichoderma polysporum	0	0	0	1 (8)					
Totals	0	0	0	23	20	18			

 TABLE 4.
 The types and numbers of fungi isolated from 20 CCA- or penta- and oil-treated Douglasfir transmission poles in service in New York, fumigated with Vapam 5 and 6 years prior to isolations.

<sup>a</sup> The number in parentheses is the isolation number as a percentage of the number of poles in the sample, for comparisons among preservatives.

<sup>b</sup> Pole sample sizes and treatments were: 8 penta- and oil-treated poles, 17 years in service and Vapam furnigated 6 years ago and 12 CCA-treated poles, 13 years in service and 9 Vapam furnigated 6 years ago and 3 poles 5 years ago. Vapam furnigations were applied as recommended by Graham and Helsing (1979).

prevalence in the treated zones coupled with fumigant effectiveness in the pentatreated poles, however, suggests primarily preservative tolerance or erratic preservative distribution and reinvasion through the CCA-treated shell.

Stored poles. – No fungi were isolated from nine penta- and oil-treated poles [four new poles and five poles stored eight years (Table 5)]. Six Cellon®-treated poles stored 7 to 10 years were also essentially fungus-free. In contrast, decay fungi were isolated from each of the four CCA-treated poles that had been stored 12 years. Three basidiomycete decayers (*Antrodia carbonica, Postia placenta,* and *Bjerkandera adusta*) were isolated from the untreated zones of four poles suggesting survival of the treatment or a deep check origin during storage. Four species of soft-rot fungi (*Phialophora malorum, Alternaria alternata, Epicoccum nigrum* and *Phialemonium dimorphosporum*) were isolated from the treated zones of the CCA-treated poles; however, no soft rot was detected microscopically as in the older CCA-treated poles in service. This indicates that soft-rot fungi can invade poles during storage and soft-rot development can be delayed until the pole is used under conditions favorable to decay (soil contact).

It is significant to note that in the stored poles, basidiomycete decayers were isolated only from untreated zones and soft-rot fungi from the outer-treated zones.

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TABLE 5. The identities, numbers and locations of fungi isolated from the approximate groundline zone of 19 treated Douglas-fir transmission poles stored in the open on racks for up to 12 years in New York.

					Number of isola	itions <sup>a</sup>			
Genus, species, or taxon of the principal fungi	Penta and oil			Cellon®			CCA		
	Outer-treated	Inner-treated	Untreated	Outer-treated	Inner-treated	Untreated	Outer-treated	Inner-treated	Untreated
			Ba	sidiomycetes					
Antrodia carbonica								1 (25)	3 (75)
Postia placenta								2 (50)	1 (25)
Bjerkandera adusta									1 (25)
			1	Microfungi					
*Phialophora malorum							1 (25)	2 (50)	
Bispora betulina								1 (25)	1 (25)
Phialemonium sp. A							2 (50)		
Phialophora sp.							1 (25)	1 (25)	
*Alternaria alternata							1 (25)		
Aureobasidium pullulans							1 (25)		
*Epicoccum nigrum							1 (25)		
*Gliocladium sp.								1 (25)	
Hormoconis resinae							1 (25)		
Moniliaceous unknown								1 (25)	
Paecilomyces variotii									1 (25)
*Phialemonium dimorphosporum							1 (25)		
Aspergillus sp.					1 (25)				
Totals	0	0	0	0	1	0	9	6	10

\* Pole sample sizes for the various treatments and storage periods were 4 CCA poles stored, 12 years; 6 Cellon poles stored between 7-10 years; and 9 penta and oil poles including 5 stored 8 years and 4 poles stored less than 1 year. The number in parentheses is the isolation number of poles in the sample for comparisons.

\* Fungi determined in this study to be soft-rot fungi.

## Soft-rot capability of selected isolates

Nine of the 12 species or taxa tested were judged to be soft-rot fungi based on weight losses exceeding 2% and/or the presence of longitudinal bore holes or erosion of the S2 zone in the cell wall. They are listed in order of decreasing soft-rot severity in Table 6. These nine soft-rot fungi represent in total more than 25% of all the isolates (417) obtained from the 163 study poles. This indicates that fungi capable of soft-rot fungi were present in highest numbers in the treated zone of CCA poles (Table 3).

Weight losses caused by the soft-rot fungi ranged from 11.0 to 2.4%. Statistical analysis indicated there are 4 or 5 levels of significant differences among weight loss means within this range (Table 6). The soft-rot fungi varied substantially in rate or degree of soft-rot capability. *Phialemonium dimorphosporum* caused the largest weight loss of the fungi tested. *Epicoccum nigrum* was judged to be a marginal soft rotter since cell-wall damage was limited to erosion of the S2 when incubated at 32 C and weight losses were barely above 2% when incubated at 24 C.

The weight loss data also indicated that optimal temperature for soft-rot development varied with the fungal species. *Phialophora malorum* and *Phialophora* sp. M had greater weight losses when incubated at 24 C while the other isolates caused greater weight losses when incubated at 32 C.

## Microscopic features of the soft rot caused by the test fungi

The microscopic features of the soft rot caused by 9 of the 12 microfungi tested are summarized in Table 7. The fungi were listed in decreasing order of weight loss (as in Table 6) to indicate the close correlation with amount of cell-wall damage in the latewood tracheids. Five of the fungi caused Type 1 damage, three both Type 1 and Type 2 damage, and one (*Epicoccum nigrum*) only Type 2 damage at the higher temperature. Cell-wall erosion and channeling (Type 2) was the major type of cell-wall damage caused by *Scytalidium lignicola*. The latter rarely formed longitudinal cavities (Type 1) in the outer S2 wall zone.

Three types of longitudinal bore holes (Type 1 soft-rot damage) were detected in the S2 of the cell walls. *Diamond* cavities were characterized by uniform conical tips with angles of approximately 30–35°. They often formed in chains and individual cavities ranged in width and length from a few microns to lengths of 80  $\mu$ m (Fig. 1-A). *Linear* cavities in contrast were narrow and long with widths ranging from 1–4  $\mu$ m and lengths up to 400  $\mu$ m (Fig. 1-B). The tip angles were also sharper and did not exceed 20°. Some linear cavities resemble longitudinal checks in the cell wall; however, hyphal remnants in the cavities and t-cell origins which can be detected readily in fiber suspensions distinguishes them. The *septate* cavities were those containing hyphal cells with conspicuous septations (Fig. 1-C). Their shapes and sizes included the ranges of the diamond and linear cavities and they may be simply cases where the penetrating hyphae remain intact. Cavity type was not fungus specific and in several cases all three types were associated with a specific fungus (Table 6). Cavity type may represent stages in bore-hole development, decay conditions, or chemical and structural differences in the various

		Incubation (	emperature C		
	33	29	24°		
Species or taxon	Mean weight loss percent and standard deviation <sup>**</sup>	Significance tests <sup>b</sup>	Mean weight loss and standard deviation <sup>a</sup>	Significance tests <sup>t</sup>	
Phialemonium dimorphosporum (ED107)	11.00 (3.47)	A, <u>A</u>	9.75 (1.25)	R, <u>A</u>	
Phialophora sp. M (ED15)	10.12 (1.29)	A, <u>A</u>	8.09 (1.88)	S, <u>B</u>	
Phialophora sp. M (ED114)	7.87 (1.22)	$\mathbf{B}, \mathbf{B}$	7.17 (1.20)	ST, B	
Phialophora fastigiata (ED142)	6.52 (0.60)	$B, \overline{B}$	6.41 (0.47)	$TU, \underline{B}$	
Phialophora richardsiae (ED117)	3.89 (0.45)	$C, \overline{CD}$	3.68 (0.93)	VWX, <u>CDE</u>	
Phialophora malorum (ED63)	3.36 (0.69)	CD, CDE	4.76 (1.99)	VW, <u>C</u>	
Phialophora malorum (ED77)	3.01 (0.26)	CD, CDEF	4.86 (1.38)	UV, $\overline{\underline{C}}$	
Scytalidium lignicola (ED105)	2.44 (0.33)	CDE, <u>DEF</u>	1.87 (0.46)	YZ, EFG	
Alternaria alternata (ED113)	2.37 (0.40)	CDE, DEF	2.82 (0.44)	XY, DEF	
Phialophora sp. (ED110)	2.36 (0.45)	CDE, DEF	3.22 (1.12)	WXY, CDE	
Epicoccum nigrum (ED115)	2.21 (0.59)	CDE, DEFG	2.39 (0.36)	XY, DEF	
Gliocladium sp. (ED83)	1.70 (0.38)	DEF, EFG	1.77 (0.61)	YZ, EFG	
Cephaloascus albidus sp. (ED48) <sup>c</sup>	1.10 (0.57)	EF, <u>FG</u>	0.71 (0.48)	Z, <u>FG</u>	
Rhinocladiella atrovirens (ED52)	1.04 (0.26)	EF, <u>FG</u>	0.92 (0.53)	Z, <u>FG</u>	
Controls	0.36 (0.21)	EF, G	0.33 (0.19)	$Z, \overline{G}$	

TABLE 6. The soft-rot capabilities (mean weight losses) of the 12 common microfungi isolated from Douglas-fir utility poles in New York as indicated by a laboratory decay test using Douglas-fir sapwood.

\* Each mean is based on 9 replications. Weight loss percent is the difference between pre- and post-oven-dry weights of the text blocks expressed as a percentage of the pre-test odw.

b Means within each temperature group and those across both temperature groups (underlined) not followed by the same letter are significantly different from each other based on the Student-Newman-Keuls test where a = 0.01 (Sokal and Rohlf 1969). <sup>c</sup> The anamorph of this isolated was *Hyalorhinocladiella* sp. ED48 was the only strain that formed asci.

	Incubation temperature								
Genus or species		32 C		24 C					
	Type of wall damage <sup>b</sup>	Cavity shape <sup>c</sup>	Cell-wall damage % (cross section) <sup>d</sup>	Type of wall damage <sup>b</sup>	Cavity shape <sup>c</sup>	Cell wall damage % (cross section) <sup>d</sup>			
Phialemonium dimorphosporum	SR1	D, S	Severe	SR1	<u>D</u> , S	Severe			
Phialophora sp. M	SR1	$\overline{L}$ , D, S	Severe	SR1	<u>I</u> , D, S	Severe			
Phialophora sp. M	SR1	<b>D</b> , S, L	Severe	SR1	$\overline{\mathbf{D}}$ , S, L	Severe			
Phialophora fastigiata	SR1	<u>D</u> , S <sup>–</sup>	Severe	SR1	<u>D</u> , s	Severe			
Phialophora richardsiae	SR1	$\overline{\mathbf{D}}$ , S	Moderate	SR1	<u>s,</u> d, l	Light			
Phialophora malorum	SR1	L	Light	SR1	S, D, Ī	Moderate			
Phialophora malorum	SR1	S, L	Moderate	SR1	S, D, L	Moderate			
Scytalidium lignicola	SR1 & 2	<u>s</u> , L <u>s</u> , L	Light	SR1 & 2	<u>s</u> , d	Light			
Alternaria alternata	SR1 & 2	<u>s</u> , l	Moderate	SR1 & 2	L	Light			
Phialophora sp.	SR1 & 2	Ī, D	Moderate	SR1 & 2	L	Light			
Epicoccum nigrum	SR2	None	None	MF2	None	None			
Gliocladium sp.	MF2	None	None	MF2	None	None			
Cephaloascus albidus	MF1	None	None	MF1	None	None			
Rhinocladiella atrovirens	MF1	None	None	<b>MF</b> 1	None	None			
Controls	None	None	None	None	None	None			

**TABLE 7.** The anatomical effects of the 12 microfungi isolated from Douglas-fir utility poles in New York on Douglas-fir sapwood after 6 months of exposure in a soft rot test.<sup>a</sup>

\* The isolate numbers of the test fungi are given in Table 6.

<sup>b</sup> Types of wall damage were defined as follows: SR1 = Type 1 soft rot-an abundance of longitudinal cavities, often in sequences, which are often conically tipped and occur primarily in the S2 portion of the secondary wall; SR2, Type 2 soft rot-localized erosion and often thinning of the S2 portion of the secondary wall initially adjacent to the S3 which also may be attacked or detached in later decay stages; Type 1 and 2 soft rot-when both are present; MF1-abundant hyphae in cell lumina but no wall damage; MF2-abundant hyphae in cell lumina with occasional transverse bore holes and/or penetration pegs. <sup>c</sup> Cavity shares are D diamond; L linear, S sentate. The predominant cavity shares is underlined.

<sup>c</sup> Cavity shapes are D, diamond; L, linear, S, septate. The predominant cavity shape is underlined. <sup>d</sup> Cell-wall damage was estimated as the percent cross-sectional area of latewood cells with cavities or erosion as seen in three randomly selected microscopic fields of the cross section at 400× magnification. The damage was ranked as follows: none; light, 10% or less; moderate, 10-50%; and severe, greater than 50%. tracheids. Combinations of microscopic features, however, readily distinguished several soft-rot fungi. *Phialophora* sp. M was characterized by numerous brown chlamydospores, appressoria and fine penetration hyphae, and linear cavities which on cross-section surfaces form a ring on the outer margin of the S2. *Alternaria alternata* was distinguished by the abundance of horizontal bore holes involving several cells in radial sequence and delamination of the S3 layer.

# Prevalence, locations and anatomical features of soft rot in poles

No soft rot was detected macroscopically in the poles.

Soft rot was detected anatomically (Type 1) in the groundline zone of 23 of the 144 Douglas-fir poles studied in transmission lines.

Soft rot was found in the outer-treated zones of 6 of 62 penta- and oil-treated poles and 4 of the 27 Cellon<sup>®</sup>-treated poles. The damage in these poles was superficial and limited to one to several outer-annual rings. The radial depth of cavity formation averaged 0.10 cm with a range of 0.005-0.22 cm. In several of these poles the soft-rot cavities were limited to several rows of surface latewood tracheids (approximately 0.005 cm). This erratic pattern may reflect poor preservative distribution.

Soft rot was found in 13 of 51 CCA-treated poles. It was often severe and in 5 poles involved the entire treated zone. In two poles the soft rot extended several cms into the untreated-heartwood zone. The radial depth of cavity formation averaged 1.05 cm with a range of 0.04–6 cm. In this group of CCA-treated poles soft rot was present also in 8 of 12 13-year-old poles which had been fumigated with Vapam 5–6 years previously.

Diamond, septate, and linear types of soft-rot cavities were detected in the poles and often all three types were present in the same soft-rot zone. Illustrations of typical soft rot in poles treated with penta and oil, Cellon<sup>®</sup>, and CCA are shown in Fig. 1.

The intensity of soft rot was highly variable in the poles and ranged from light (less than 10% cell-wall damage) to moderate (40–50% cell-wall damage) in each preservative-service age group where soft rot was found. Severe soft rot (more than 75% cell-wall damage) was detected only in several of the 15-year-old CCA-treated poles.

No soft rot was detected in 19 stored poles although fungi capable of causing soft rot were isolated from several stored poles.

## DISCUSSION AND SUMMARY

A large and diverse group of wood-inhabiting fungi is associated with the decay of Douglas-fir poles in the groundline zone. An estimated 70 species or genera were obtained from 144 of the 163 poles sampled and 20 appeared 4 times or more and are presumed to play a possible direct or indirect role in decay development. These 20 genera or species represented nearly 60% of the 417 isolates obtained.

Microfungi predominated over Basidiomycetes in the groundline zone of the poles and comprised nearly 85% of all isolates. They were obtained most commonly from the treated zones of the poles and in greatest abundance from the older CCA-treated poles.

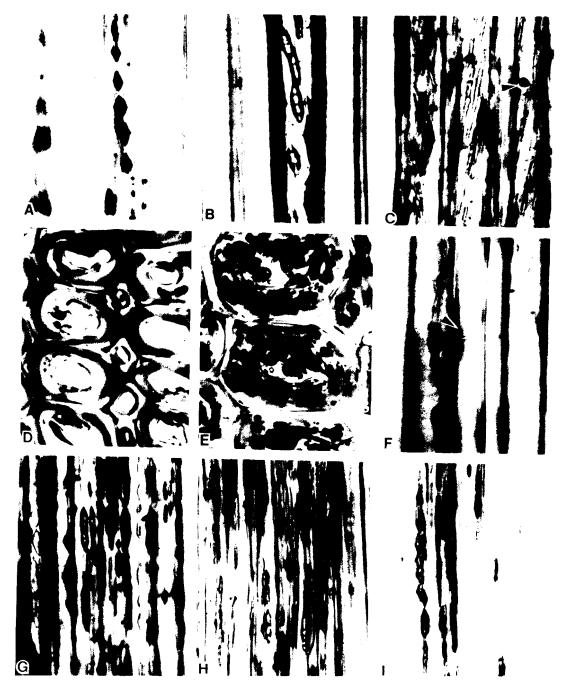


FIG. 1. A-F. The three types of longitudinal bore holes formed by the test microfungi in Douglasfir sapwood test blocks. A. *Diamond* cavities associated with *Phialemonium dimorphosporum*, radial section,  $500 \times$ ; B. *Septate* cavities associated with *Phialemonium dimorphosporum*, radial section,  $400 \times$ ; and C. *Linear* cavities associated with *Phialophora* sp. M, radial section,  $550 \times$ . The arrow indicates one of the large brown chlamydospores associated with this fungus; D. Types 1 and 2 softrot damage associated with *Alternaria alternata*, transverse section,  $600 \times$ ; E. Type 1 cavities in a ring

Decay tests established that 9 of the 12 genera or species of microfungi tested were fungi capable of producing soft-rot deterioration. The three most prevalent soft-rot fungi were *Phialemonium dimorphosporum*, *Scytalidium lignicola*, and *Phialophora mutabilis*. *Phialemonium dimorphosporum* is a recently described anamorph genus (Gams and McGinnis 1983) and reported here for the first time known to us as a common inhabitant of Douglas-fir poles and capable of causing serious soft-rot damage in wood. These 9 fungi capable of causing soft-rot damage comprised 25% of all the isolates from the groundline zone of the poles. Probably the number is higher since many fungi in the miscellaneous and general groups were untested and may contain additional soft-rot fungi.

Four species of *Phialophora* were among the frequently isolated fungi, which agrees with prior reports from northern Europe and Australia (Nilsson and Henningsson 1978; Daniel and Nilsson 1988) on the importance of this group in the decay of utility poles.

These data indicate that soft-rot fungi are an important component of the woodinhabiting fungi that invade treated poles of Douglas-fir. Their presence in the outer-treated zones of poles, with their established decay capacities, suggests an important role in early decay development.

The principal basidiomycetes isolated were *Antrodia carbonica* and *Postia placenta*. They were isolated primarily from the untreated zones of the CCA-treated poles both in storage and service. They were generally associated with localized pockets of an advanced brown-cubical rot. Five of the six white-rot basidiomycetes were isolated from the treated zones of the penta-treated poles. These results agree generally with prior reports on the important decay fungi in Douglas-fir poles (Eslyn 1970; Zabel et al. 1980; Morrell et al. 1987).

Two probable origins are proposed for these wood-inhabiting fungi based on isolate location and prevalence. Deep checks developing into the untreated heartwood are the likely colonization routes for the fungi obtained primarily from the untreated zones of the poles. The colonization may occur in the storage yard or the pole in service when deep checks develop upon drying below the fiber saturation point. A related origin may be check invasions during seasoning and failure of the preservative treatment to sterilize the inner regions of the pole. Shallow checks into the lower retention zones of the inner-treated sapwood are likely entry points for the fungi obtained primarily from the treated zones. Many of these fungi probably could be placed among the stress-tolerant wood colonizers described by Rayner and Boddy (1988). The soft-rot fungi with the single exception of Scytalidium lignicola were obtained primarily from the treated zones of the CCAtreated poles. Scytalidium lignicola was isolated with one exception from the penta-treated poles. Another case of close fungal association with a preservative treatment was the *Hvalorhinocladiella* sp. which was isolated primarily from the treated zones of the penta- and oil-treated poles. This is in agreement with a report

pattern associated with *Phialophora* sp. M, transverse section,  $800 \times$ ; and F. A longitudinal cavity developing from a T-cell in *A. alternata*, 1,200 ×.

G–I. Soft-rot damage in Douglas-fir utility poles. G. Diamond cavities in a 13-year-old CCA-treated pole fumigated with Vapam in 1979, radial section,  $350 \times$ ; H. Septate cavities in a 15-year-old CCA-treated pole, radial section,  $350 \times$ ; and I. Diamond cavities in an 8-year-old pole treated with penta and oil, radial section  $350 \times$ .

by Polishook (1982) on the common association of this fungus with penta- and oil-treated southern pine poles. This fungus was an early colonizer and isolated less frequently in older poles (15–18 years in service).

The detection of soft rot to date has been based primarily on anatomical evidence, i.e., the formation of longitudinal bore holes in the S2 of the secondary wall (Type 1) or the thinning and erosion of the secondary wall (Type 2). While Type 1 soft rot can be readily identified and quantified microscopically, Type 2 soft rot is difficult to judge, particularly in conifers, where the early soft-rot attack is primarily wall thinning in the S-2 zone (Nilsson 1973). Previous studies of soft rot in southern yellow pine have shown that at comparable levels of cell-wall damage to the most severe ones observed in this study *Scytalidium lignicola*, *Alternaria alternata*, and *Epicoccum nigrum* caused strength losses in the toughness of test beams of 25.8, 19.5, and 18.2%, respectively (Meyer et al. 1988). For these reasons we believe that weight losses from decay studies may be a more reliable way to detect and quantify the soft-rot capability of a non-basidiomycete since longitudinal cavities are often sporadic in the sections and cell-wall erosion is difficult to quantify.

Typical soft rot, as characterized by a pattern of rectangular-shaped shrinkage cracks, was not seen visually on the pole surfaces exposed in the groundline zone. Some surface softness was detected in several shallow pockets on primarily the older CCA-treated poles.

The difficulty of visually detecting early soft rot indicates a need for periodic cultural and/or anatomical checking to determine its presence for decisions on the timing of remedial groundline treatments.

Soft rot was detected anatomically (Type 1), in 23 of the 144 transmission poles studied in service lines. It was limited to several outer rings in the penta-treated poles and judged to be minor. Soft rot was often severe in the older CCA-treated poles and in some cases penetrated all of the treated zone and extended several centimeters into the inner-untreated zones. This probably reflects the tolerance of soft-rot fungi to CCA-type preservatives and has been reported previously (Henningsson et al. 1975; Dickinson et al. 1976; Levy 1978; Leightley 1980).

Soft rot was detected both anatomically and culturally (soft-rot fungi isolated) in 8 of 12 13-year-old CCA-treated poles which had been fumigated with Vapam 5-6 years previously. Whether the soft-rot damage occurred prior to the fumigation or resulted from fungal invasion after fumigation is unknown. However, the isolation of several species of soft-rot fungi after fumigation from the soft-rot zones suggests that preservative tolerant soft-rot fungi invaded the poles and caused the rot. This suggests that a shorter fumigant retreatment cycle may be necessary for some CCA-treated poles and supplementary groundline treatments considered. No fungi were isolated from 8 penta-treated poles, which had been in service for 17 years and fumigated 6 years previously with Vapam.

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