AN ABSTRACT OF THE THESIS OF

Paul Przybylowicz for the degree of

Doctor of Philosophy in Botany and Plant Pathology

presented on 4 February, 1985.

Title: Establishment of Poria carbonica in Wood and Colonization by Basidiomycetes of Douglas-fir Utility Poles During Air Seasoning

Abstract approved: Redacted for privacy

M. E. Corden

Influence of temperature and wood moisture content on spore germination and colonization of Douglas-fir (Pseudotsuga menziesii) heartwood by Poria carbonica basidiospores and chlamydospores was studied. Both spore types germinated and colonized wood at moisture contents above the fiber saturation point, but not below. Germination and wood colonization occurred at 22°C or 30° C, but not at 5 or 35°C. Chlamydospores germinated at 5 and 35°C, but were unable to colonize the wood.

Basidiomycete colonization of air-seasoning Douglas-fir poles in the Pacific Northwest was studied during a 2 year period. Sterilized pole sections (2 and 4 ft long) were exposed for 3-month intervals at four widely separated sites. Basidiomycetes were isolated from the pole sections by removing wood increment cores, placing the cores into nutrient medium, and then culturing fungi that grew from the wood.

Twenty-five basidiomycetes were identified from the pole sections including six monokaryons (identified by successful matings with known monokaryotic isolates). Basidiomycete isolation frequencies were generally higher during the winter months, and were particularly high in the period November, 1981 to January, 1982.

The isolation frequency and distribution of basidiomycetes in 1540 air-seasoning Douglas-fir poles from 24 yards in the Pacific Northwest were determined by removing 14 increment cores from each pole. The poles ranged from freshly cut to those seasoned over 25 months.

Thirty basidiomycete species including 11 with monokaryons were isolated from the air-seasoning poles. The frequency of poles colonized by decay fungi increased from 35% following pole felling to about 90% after 18 months of air seasoning. Poria carbonica and P. placenta, the primary decay fungi in Douglas-fir poles in service, were among the most prevalent basidiomycetes after 25 months of air seasoning. These species generally were isolated from the heartwood where they may survive some preservative treatments to initiate internal decay in poles in service. Limiting the air-seasoning period to 1 year and sterilizing air-seasoned Douglas-fir poles, especially transmission poles, during preservative treatment should markedly decrease decay in poles in service.

Establishment of *Poria carbonica* in Wood and Colonization by Basidiomycetes of Douglas-Fir Utility Poles During Air Seasoning

by

Paul Przybylowicz

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirement for the
degree of

Doctor of Philosophy

Completed February 4, 1985

Commencement June, 1985

APPROVED:

Redacted for privacy

Professor of Botany and Plant Pathology in charge of major

Redacted for privacy

Head of Department of Botany and Plant Pathology

Redacted for privacy

Dean of Graduate School

Date thesis is presented 4 February, 1985

Word processed by Paul Przybylowicz

ACKNOWLEDGEMENTS

I would like to thank my associates involved in this project for their support and help. My sincerest thanks go to Dr. Bradley Kropp, who spent countless hours identifying many of the fungi isolated from the poles, to Dr. M. E. Corden for his guidance and assistance, and to Helen Gehring who assisted in many experiments. I would also like to thank Camille Sexton for her help and support, Paul Hessburg for his thoughtful comments, Jeremy Wilson for his enthusiasm, guidance and cheerful answers on computer analysis at all hours, and Bob Brenne for his help in the data analysis.

My thanks also extend to:

The U.S. Forest Products Laboratory and Dr. W. Eslyn in Madison, WI, the Bonneville Power Administration, Portland General Electric Co., and Empire State Electric Energy Research Corp. for major funding of this research.

Prof. Robert Graham, Gary Cantara and Mark Newbill of the Forest Products Department of Oregon State University for their invaluable technical assistance,

and all the undergraduate student workers who helped out especially Ramona Vincent, Jay Sexton and Saundra Anderson.

Last, but not least, my thanks to Barbara Lee for her support, help, and patience.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

Establishment of Poria carbonica in Wood and Colonization by Basidiomycetes of Douglas-fir Utility Poles During Air Seasoning

INTRODUCTION

Douglas-fir (Pseudotsuga menziesii) is widely used for distribution poles up to about 45 feet long and transmission poles that may be as long as 120 feet. When seasoned and pressure-treated with a preservative in conformance to national standards (2), Douglas-fir poles provide long service, usually 30 years or more (25). However, in transmission poles, a large core of untreated heartwood remains. Because large poles cannot be seasoned to the low moisture contents they generally reach in service, some poles develop deep seasoning checks (26). These checks expose untreated heartwood which is generally believed to be the site of successful colony initiation by decay fungi (10,16,27,31,104). However, decay fungi present in the wood before preservative treatment may survive some low-temperature treatments to cause internal decay of poles in service (16,104).

Freshly cut poles must be partially dried prior to preservative treatment to obtain adequate penetration by preservatives (36). Kiln drying and Boulton drying (heating poles in creosote or pentachlorophenol in heavy oil under vacuum) were widely used to dry poles. In recent years, there has been a shift toward air-seasoning

of Douglas-fir poles because of high energy and oil costs and to reduce wood strength loss due to heating. However, the time required to adequately air season large pole may allow decay fungi to become established in the wood.

The relatively high temperatures (usually above 94°C) and long heating times used during kiln and Boulton drying are generally sufficient to kill decay fungi inside even large poles (28,30,44). Air seasoning reduces the heating time required to pressure-treat poles, but the reductions in heating times and temperatures may allow decay fungi, inside poles, to survive.

Pentachlorophenol in heavy oil, is widely used as a pole preservative. The high cost of oil and the desire for a clean pole have caused a shift toward non-oily preservatives. During treatment with water-borne and other clean preservatives (e.g. pentachlorophenol in liquified petroleum gas, mineral spirits or methylene chloride) temperatures generally are low (36,43) and may allow decay fungi in the heartwood to survive treatment. The use of pentachlorophenol may be limited in the near future because of its toxicity and potential for environmental contamination (25,31,36).

During a study of utility pole decay in the Northwest, an unidentified basidiomycete was isolated from ponderosa pine poles in one pole line in Montana (16). This fungus, usually found in living trees, reportedly had not been

isolated from any other wood products. It was suggested that the poles were from heavily infected trees prior to cutting, and that the fungus survived preservative treatment to cause decay in the poles in service.

A recent study of Douglas-fir poles in service in the Northeast (104) suggests that decay fungi may survive treatment with chromated copper arsenate (CCA) and continue to decay poles in service. Poria carbonica and P. placenta were found in 72% of the Douglas-fir poles treated with CCA, but only in 15% of the poles treated with pentachlorophenol in heavy oil. P. carbonica is uncommon in the Northeast, so the high isolation frequency suggests that the poles were colonized by P. carbonica during air seasoning in western yards, where this fungus is prevalent.

Interest in pretreatment decay was stimulated by Taylor (94) who proposed strict specifications which required sterilizing poles and that Douglas-fir poles were to be preservative treated within 3 months of cutting, which eliminated air seasoning. He also suggested that Douglas-fir poles become infested by decay fungi within 3 to 6 months of cutting and that significant decay occurs within 6 to 12 months. However, none of these suggestions were supported by research on colonization of air-seasoning Douglas-fir poles.

The major fungi associated with decay in Douglas-fir poles in service have been documented (12,14,16,104), but relatively little is known about the incidence of decay fungi in poles during air seasoning. The purpose of this study was to develop an understanding of the process of colonization of air-seasoning Douglas-fir poles by decay fungi and to determine the extent of this colonization.

Understanding spore germination is critical to developing effective control measures. During germination decay fungi are not yet established and thus are most susceptible to inhibitory chemicals (84). I developed a method for establishing decay fungi in wood using spores, under conditions similar to those in seasoning checks in poles in service. This method was used to investigate the effects of wood moisture content and temperature on Poria carbonica spore germination (Chapter 1), and may allow more realistic, rapid testing of new chemicals to prevent wood colonization.

Pole colonization by decay fungi involves interactions between the substrate (poles), the environment, and the decay fungi. Successful colonization occurs when a fungal propagule lands on a pole and environmental conditions are favorable for its germination and growth. We studied colonization of poles periodically during the year using sterilized pole sections as spore traps (Chapter 2). Temperature and precipitation were examined as predictive

factors for periods with high potential for wood colonization. Specific management practices during these periods could reduce pole decay.

To determine the extent of pretreatment decay, we sampled Douglas-fir poles in air-seasoning yards in the Pacific Northwest which included northwest California, western Oregon, and western Washington. This study (Chapter 3), identified the decay fungi involved, and estimated their frequency in poles during air seasoning.

GENERAL METHODS

CULTURE MEDIA

Malt-extract agar is commonly used for culturing wood decay fungi (4,16,31,63). The following culture media, based on malt or potato dextrose agar (PDA), were used during these studies. Malt medium I was used for isolation of decay fungi from wood and to grow *P. carbonica* cultures for chlamydospore isolation. Malt medium II was used to maintain cultures after isolation and, with the addition of Douglas-fir heartwood blocks, for *P. carbonica* basidiospore production. Acid-benomyl-malt medium was used to isolate pure cultures of potential decay fungi from cultures overgrown by fast-growing fungal and bacterial contaminants. Acid PDA was used to isolate *P. carbonica* from inoculated wood, because the low pH favored *P. carbonica* growth and inhibited other fungi and bacteria growing from the wood.

Media (for 1 liter)

Malt extract medium I	25 g	malt extract
	8 g	agar
Malt extract medium II	12.5 g	malt extract
	10 g	agar
Acid benomyl malt medium	25 g	malt extract
	8 g	agar
	10 mg	benomyl
	0.075 ml	lactic acid (85%)

Acid potato dextrose agar (PDA)	200 g	potatoes
	20 g	dextrose
	10 g	agar
	0.075 ml	lactic acid (85%)

The lactic acid and benomyl were added after cooling the autoclaved media, just prior to pouring into petri plates.

ISOLATION AND IDENTIFICATION OF DECAY FUNGI

Poles were sampled by removing wood cores (3 mm dia X 15 cm long) with an increment borer. The cores were placed singly in labelled, plastic, drinking straws which were stapled shut and kept cool to minimize microbial growth. The cores were returned to the laboratory for isolation of decay fungi. Each core was cut into pieces about 1 inch long, flamed to surface sterilize, and then partially embedded in malt extract medium I in petri plates. Decay fungi growing from the core sections often grew along the wood surface before growing onto the culture medium. Thus, separation of the core sections prevented fungi in one section from overgrowing the entire core.

The cultures were first examined about 2 weeks after plating when locations of contaminants and possible decay fungi were marked on the petri plate and recorded. The origin of fungal isolates at later examinations could not always be determined because many cultures were overgrown by fast-growing fungi.

After a minimum of 1 month, the cultures were examined a second time, and possible decay fungi were examined microscopically. These fungi, sub-cultured on malt medium I for identification, generally fell into two major groups: (i) basidiomycetes with clamp connections on the mycelium, and (ii) suspect fungi which lacked clamp connections, but possessed other features characteristic of basidiomycetes (e.g. fiber hyphae, hyphal size and appearance, white cottony mycelium). Many suspect fungi were later identified as monokaryotic basidiomycetes. Possible decay fungi overgrown by fast-growing imperfect fungi were often obtained in pure culture by streaking onto acid-benomyl-malt medium. The presence of non-basidiomycetous fungi was recorded but these fungi were not studied further.

Dr. Bradley Kropp, Oregon State University, identified most of the decay fungi using published keys (63,90). When keys failed to provide identification, fungi with clamp connections were compared to known cultures obtained from other researchers or by culturing from identified sporophores. Unidentified suspect fungi were paired with monokaryotic or dikaryotic isolates of known fungi. The formation of clamp connections indicated that the isolates were the same fungal species. Some isolates were also sent to the Center for Forest Mycology Research, U.S.

Forest Products Laboratory, Madison, Wisconsin, for identification and verification.

DATA ORGANIZATION AND ANALYSIS

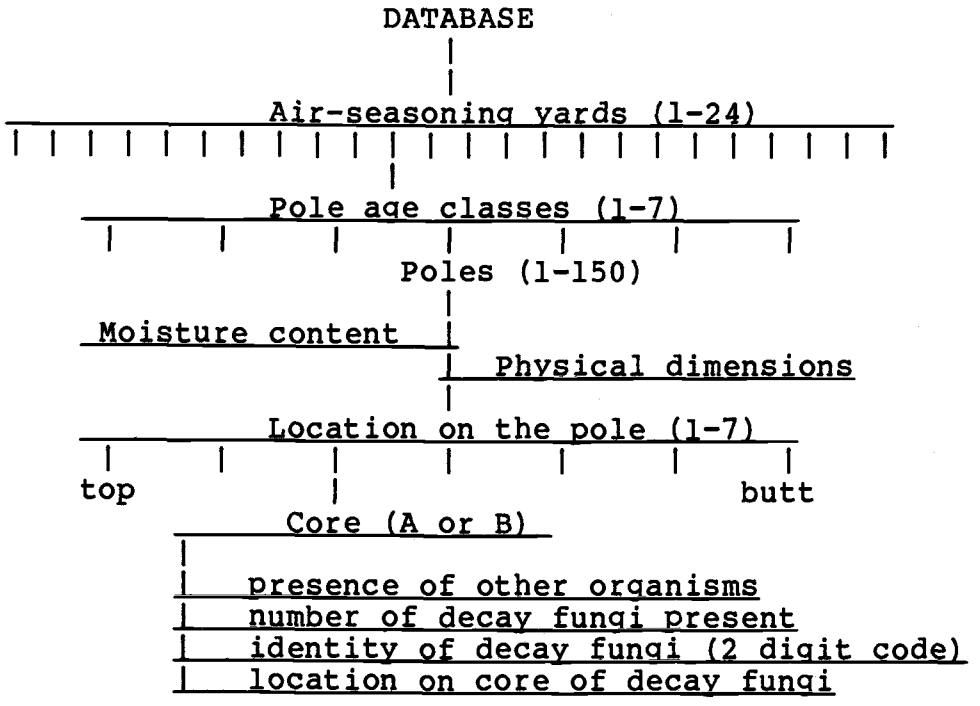
The large amount of data from the field studies (over 32,000 lines) were entered into an Apple IIe microcomputer using the Apple Pascal editor, and then transferred via a modem to a Cyber 170/720 mainframe computer for sorting and storage. This minimized the use of expensive mainframe time for data entry. However, because of recent increases in the memory capacity of microcomputers, this type of analysis could now be accomplished entirely on a microcomputer.

The data were organized into two databases using the Scientific Information Retrieval Service (SIR) database software. The SIR software, designed to handle large amounts of data, was not "user friendly" and consequently was difficult to use.

Large databases are organized into a hierarchical structure to maintain the relationships between the data and then into records, lines of data that are differentiated by "sort keys" (variables which contain unique values for each record). Records are separated into groups by common sort keys and share the sort keys of higher levels of the hierarchy. A clear definition of the desired analysis is necessary to choose sort keys that allow efficient data sorting, whereas poorly chosen sort

keys will increase the programming and computer time required for analysis.

For example, the air-seasoning study was designed to determine the frequency and distribution of decay fungi in air-seasoning Douglas-fir poles in the Pacific Northwest. The desired analysis required sorting by pole age classes, yard location, and core position on the poles which were specified as sort keys. Thus, the data were organized into single lines for each core containing codes for yard location, sampling date, pole number, position on the pole where core was removed, and number of basidiomycetes isolated from the core and their location. By specifying the desired sort keys, the data could be sorted in many different ways to produce tables (e.g. the number of poles per yard containing decay fungi or the number of cores containing Poria carbonica in the third age class). Additional records contained information on the physical dimensions and moisture content (based on oven-dry weight) of each pole and shared the same pole, age class, and yard identifiers with the core records. This type of hierarchical approach is basic to handling large amounts of data with a computer. The air-seasoning database can be represented as follows.


DATABASE
Air-seasoning yards (1-24)
Pole age classes (1-7)
Poles (1-150)
Moisture content
Physical dimensions
Location on the pole (1-7)
top
butt
Core (A or B)
presence of other organisms
number of decay fungi present
identity of decay fungi (2 digit code)
location on core of decay fungi

CHAPTER 1

The Influence of Temperature and Wood Moisture Content on Spore Germination and Colonization of Douglas-fir Heartwood by Poria carbonica

INTRODUCTION AND LITERATURE REVIEW

It is generally accepted that most basidiomycetes are dispersed by wind-borne spores (18), although colonization by direct hyphal growth from decayed wood may be important in localized spread. Seasoning checks and exposed endgrain in poles are the most likely sites of successful basidiomycete colonization because they trap dust and water (16,27,104). Many environmental factors influence spore germination and wood colonization by decay fungi, but only temperature and wood moisture content were examined during this study because these factors are probably the most important under natural conditions.

Poria carbonica causes most of the decay in Douglas-fir utility poles (16,104); however, little is known about its dissemination and establishment in wood. P. carbonica produces basidiospores, chlamydospores and conidia, which could initiate decay. Basidiospores are sexually produced in a specialized sporophore and, when mature, are dropped into the air where they can be transported long distances by wind (18,39,44,101). Thus, basidiospores are the most likely dispersal spores. Chlamydospores and conidia are produced asexually throughout the mycelium and, although they appear similar, chlamydospores have thicker walls

(63) which are more resistant to environmental extremes. Poles could be colonized by these asexual spores associated with soil or dust particles, or by direct contact with soil or decaying wood during logging, yarding, and storage.

Studying wood colonization by P. carbonica required methods for isolating the different spore types in suitable quantities. Chlamydospores and conidia are readily produced in culture, but must be separated from the fungal mycelium. Basidiospores can be collected from naturally occurring sporophores or from sporophores produced in culture. Spores collected in the field often have bacterial or yeast cells associated with them. Although these contaminants may be important in wood colonization, they introduce unwanted variables into controlled laboratory experiments. Consequently, basidiospores obtained from sporophores produced in culture are more desirable.

Sporophore and basidiospore production in culture

Many methods have been developed to induce cultures of wood-decay fungi to produce sporophores in culture, but none have dealt with P. carbonica. An early study which examined the ability of wood-rotting fungi to produce sporophores on different artificial media (49), concluded that malt agar was best and that light generally was

required. Sporophores from 16 genera, including 42 species of decay fungi were obtained, but only the Agaricaceae produced typical sporophores.

Fructification of many wood-decaying basidiomycetes was achieved in diffuse daylight on sawdust-containing media (17) or water-saturated sterile sawdust amended with corn meal, bone meal, potato starch, saccharose, or wood ashes (3,4). However, providing adequate moisture after the onset of sporophore formation was difficult, and sporophores generally were atypical. Sterile wood blocks added to the cultures and incubation under high humidity (87,93) resulted in more typical sporophores.

Normal sporophores of Phellinus weirii (48), Polyporous arcularius (19), and Poria ambigua (75) were produced on 2% malt agar. P. weirii and P. ambigua required light for sporophore formation. Gloeophyllum saeparium (91) produced typical sporophores only on wood blocks that were partially dried following colonization. The general objective of these studies was to produce sporophores from mycelial cultures for identification. Although spore production and viability were not reported, aseptic collection of spores produced by these methods would have been difficult.

To obtain basidiospores, sporophores of wood-decaying basidiomycetes were produced on 2% malt agar in inverted petri-plate cultures (58,59,81,96) which allowed aseptic

collection of spores from the plate covers. Gloeophyllum saepiarium, G. trabeum, or Fomitopsis roseus were grown on malt extract medium, incubated until the colony diameter reached about 3 cm and then inverted at room temperature under diffuse daylight. Sporophores usually were produced about 3 weeks after inversion.

Observation of germinating spores

The influence of environmental factors such as CO_2 concentration, pH, temperature, and relative humidity on spore germination of wood-decaying basidiomycetes on agar media have been studied (23,38,54,59,60,97). Germinating spores are easily observed on agar media, but the results of these studies must be viewed in light of the large physical and nutritional differences between nutrient agar media and wood.

Several approaches have been used to observe spore germination on wood. The contact-agar-block method (82,83,84) consisted of fusing a small water-agar block to the wood, placing spores on the agar medium and observing spore germination. Another approach (52) was to place spores on a dialysis membrane which was then placed on the wood surface, and finally removed for observation of spore germination. While these methods allowed soluble substances from the wood to diffuse to the spores, they were unsatisfactory for this study because an artificial

barrier was between the spores and the wood, and it prevented evaluation of subsequent colonization.

Direct observations have been made of stained, germinated, decay-fungus spores on thin wood-sections (59,83,96,106). These methods were unsuitable for this study because the wood sections were too small to accurately control their moisture content. Spores can be observed directly on wood surfaces using scanning electron microscopy (SEM), but a simpler technique was used for most of the studies reported here.

Objectives

Poria carbonica and P. placenta have been used as test fungi in wood preservative and fumigant evaluation studies (11,29,98). The inoculum used in these tests generally consists of a relatively large section of agar culture medium with actively growing mycelium of the test fungus. Because of the large amounts of inoculum used and the food reserves in the culture medium, decay fungi may overwhelm wood preservatives in laboratory tests. However, these chemicals may be effective in the field where colonization is generally initiated by single spores. The ability to initiate wood colonization from single spores in controlled laboratory experiments could result in a closer correlation between laboratory and field evaluations of potential wood preservatives.

Investigating the establishment of P. carbonica in

Douglas-fir heartwood by spores and the influence of temperature and wood moisture content on this process required methods for: isolating the spores, placing them on wood, and observing germination. Thus, the objectives of this study were: (i) to isolate and collect P. carbonica chlamydospores and conidia from growing cultures, (ii) to induce P. carbonica to form basidiospores in culture, (iii) to develop techniques for observing spore germination on wood blocks large enough to accurately control the wood moisture content, (iv) to determine if individual propagules of P. carbonica are capable of establishing colonies in wood, and (v) to examine the influence of temperature and wood moisture content on the establishment of P. carbonica in wood.

MATERIALS AND METHODS

Isolation of soil particles containing viable Poria carbonica

Soil particles containing viable P. carbonica may act as short-range dispersal propagules. To obtain these particles, a Jory-series soil was collected from MacDonald Forest, Corvallis, OR. The soil was obtained from no deeper than 25 cm and was sieved through a screen with openings of 8 mm to remove most of the large organic debris. The soil was brought to field capacity with distilled water and about 250 cc per flask were placed in ten 500 ml Erlenmeyer flasks. The flasks then were

plugged with cotton, and autoclaved for 40 min at 15 lb. Soil from several, randomly chosen flasks was combined, weighed, oven dried (110°C for 24 hr) and reweighed to calculate moisture content, which was between 41 and 49%. The soil in the remaining flasks was inoculated with a suspension of P. carbonica hyphal fragments and asexual spores and incubated at 30°C for 1 month. During incubation, the flasks were periodically shaken to distribute the decay fungus evenly throughout the soil.

After incubation, the flasks of soil were placed in a forced-air oven at 28°C for 1 week to dry the soil. Then the soil was sieved through a series of six screens with openings ranging from 0.99 to 0.038 mm. Small amounts of each particle size (tip of a small spatula) were placed on Douglas-fir heartwood blocks (2 X 2 X 1 cm) which had been saturated with sterile distilled water under vacuum for 30 min and autoclaved for 20 min at 15 lb. The inoculated blocks were incubated at 22°C for 1 month. Three replicates of 12 blocks per particle size were conducted.

To determine successful colonization by P. carbonica, each block was split aseptically, and four small wood chips from the block interior were removed and partially embedded in acid PDA. Two weeks later, the chips were examined for growth of the decay fungus.

Isolation of chlamydospores

Chlamydospores and conidia of P. carbonica are about the same size and shape (63), and are differentiated by the thicker walls of the chlamydospores. Microscopic examination of the isolate used revealed both types of spores were present in the mycelium, although most were chlamydospores. The following technique does not differentiate between the two spore types, thus the chlamydospore suspensions contained some conidia.

Chlamydospores were isolated from mycelial mats of P. carbonica grown on malt extract medium I (see general methods for media details). Disks of water-permeable cellophane were cut to fit inside the petri plates. The disks were autoclaved and placed on the culture medium surface prior to inoculation with mycelium. This permitted rapid removal of the mycelium from the culture without removing any of the medium. Culture medium in the mycelial suspension was undesirable because it clogged the filter paper used to separate the spores from the mycelium.

The cultures were incubated in the dark at 30°C until the cellophane was covered with mycelium. Then the mycelium was aseptically scraped off the cellophane and placed in a screw top test tube, containing 25 ml of sterile distilled water and about 3 cm of broken glass in

the bottom. Rapid shaking of the tube resulted in the mycelium being cut into small fragments by the glass pieces. The resulting suspension was filtered through four layers of cheesecloth and a Whatman #4 filter paper. The filtrate was centrifuged at 3000 g and 5°C for 15 min and the supernant was discarded. The pellet was resuspended in 2 ml of sterile distilled water which gave a chlamydospore suspension relatively free of hyphal fragments. The chlamydospore concentration was adjusted using a haemacytometer to about 10^6 spores per ml. Spore suspensions from several plates were combined, centrifuged and resuspended when the initial spore concentration was too low.

Isolation of basidiospores

Because P. carbonica does not readily produce basidiospores in culture, different methods to induce sporulation were tried, including: nutritional shifts from high sugar, high nitrogen, to low sugar, low nitrogen; near-ultraviolet light treatments; rapid temperature changes; different culture media; and addition of wood to the culture medium. Sporophores were induced by all these methods, but viable spores were produced reliably only when wood was added to the culture medium.

Based on these preliminary experiments, a method was developed whereby basidiospores were collected from sporophores produced in petri plates on malt medium II,

which had two small Douglas-fir heartwood blocks (2 X 1 X 0.5 cm) added. The blocks were water-saturated under vacuum, autoclaved, and placed in the culture medium before it solidified. The medium was inoculated with P. carbonica mycelium, incubated at 30°C in the dark until colony diameters reached about 2-4 cm, and then the plates were inverted under diffuse light conditions at 20-22°C. Basidiospores, collected on the petri-plate lids, were usually produced 3 to 4 weeks after inversion. The lids were changed 24 hours prior to spore use to ensure collection of fresh spores. Spore suspensions were prepared by washing the spores off the cover with 1 ml of sterile distilled water, and adjusting the concentration, using a haemacytometer, to 1-4 X 10^6 spores per ml.

Observation of spores on wood, the "spore sandwich"

Due to their small size (2-2.5 X 5-5.5 μm), individual P. carbonica basidiospores are difficult to observe on wood surfaces. Several different techniques were tried while developing a successful method. For example, basidiospores were placed on a radial face of a Douglas-fir wood cube (1 cm^3) that had been planed with a sliding microtome. The spores were stained on the wood with cotton or trypan blue, then observed with epi-illumination through a microscope. Because of the relatively large diameter of the tracheids (about 35-45 μm) (66), the wood

surface was microscopically uneven. Most of the spores below the upper edges of the tracheids could not be observed. The reflected light from water on the wood surface also made spore observation difficult.

In the final method, thin, radial, Douglas-fir heartwood sections (8 X 8 mm X 60 μm) were cut with a sliding microtome and placed between two glass slides to hold them flat. The slides containing the wood sections were placed in petri plates and autoclaved to sterilize the wood. The sections were removed from the slides, placed on sterile filter paper in petri plates, and inoculated with 25 μl of a spore suspension. Excess water was wicked through the sections by the filter paper, leaving the spores deposited on the wood surfaces. A second section was then placed on top, forming a "spore sandwich" with the spores between the two sections. The "spore sandwiches" were removed from the petri plates and placed between the halves of a Douglas-fir block (2 X 1 X 1 cm, the 1 X 1 cm face was a radial face), which were secured with a rubber band. The block cultures were then incubated in the desired temperature or humidity chamber for 4 days.

Initially, the wood sections were inoculated by placing them in a sterile petri dish, and suspending a petri-plate culture with a sporulating sporophore above the sections. Both plates were placed in a dessicator

over distilled water to maintain high humidity. The top plate was rotated slowly relative to the bottom plate by means of a mechanical turntable. This ensured even spore distribution without introducing water with the spores. Unfortunately, P. carbonica produced insufficient spores for this method to be successful. Spore densities on the sections were not high enough to observe 100 spores per section. Other basidiomycetes used in trying this technique, e.g. Fomitopsis cajanderi, sporulated heavily and produced uniform, dense spore deposits on the wood sections.

Following the incubation period, the "spore sandwiches" were removed from between the block halves, and the wood sections were peeled apart. Each section was carefully placed, inner side up, on a small drop of stain (0.05% trypan blue in lactophenol) on a microscope slide; the stain colored the spores without washing them from the sections. Examination of both sections allowed an equal chance for each spore to be counted. Spores were scored as germinated when the germ tube length exceeded the spore diameter.

Determination of wood colonization initiated by spores

One half of the blocks containing "spore sandwiches" were incubated for 1 month, after which aseptic isolations were made 1 mm in from the split face on each block half

to determine colonization success of the decay fungus. Wood chips were removed, placed into acid PDA, and observed after incubating 1 month at 22°C for growth of *P. carbonica*.

Preparation of wood sections for scanning electron microscopy

Germinating basidiospores on thin wood-sections from the "spore sandwich" were observed at 2, 3, and 4 days after inoculation with scanning electron microscopy (SEM). The inoculated wood sections were freeze-dried, then coated with gold-palladium (60:40) under a pressure of 1 X 10^{-5} torr in a Varian-Mikros VE-10 vacuum evaporator. Specimens were examined with an Amray 1000A scanning electron microscope operating at 20 KV accelerating potential with a 100A beam. Photographs were made with Polaroid type 55 P/N film.

Experimental design for the influence of wood moisture content on spore germination on wood

Thin wood-sections alone were unsatisfactory for investigating the influence of moisture content on spore germination because it was impossible to adjust and maintain the sections at a desired moisture content. However, blocks enclosing the sections provided a volume of wood large enough to accurately achieve the desired wood moisture content.

To study the influence of wood moisture content on spore germination, the Douglas-fir heartwood blocks used to enclose the "spore sandwiches" were oven dried (24 hr at 110°C) and weighed along with a rubber band. The blocks were then split with a razor blade, and banded together with the rubber band.

Three moisture levels were used: below, at, and above the fiber saturation point (FSP) which is about 26 to 32% (based on oven-dry weight) in Douglas-fir. To produce wood below the FSP, blocks were placed on glass supports over a saturated solution of monobasic ammonium phosphate, in sealed jars, which gave a relative humidity of about 93% at 22°C (102). Wood at the FSP was obtained by placing blocks over distilled water, while blocks above the FSP were saturated with water under vacuum for several hours, then placed over distilled water. The jars containing the blocks were autoclaved, sealed, and then held at 22°C for at least 2 weeks before use.

The blocks were weighed before inserting the "spore sandwich" and after incubation to obtain an average wood moisture content for the incubation period. The humidity chambers containing the block cultures were placed inside plastic crispers and incubated in a growth chamber at 22° C. Each spore type was tested in three replicates of 18 blocks, six blocks per wood moisture level.

Experimental design for the influence of temperature on spore germination on wood

The "spore sandwich" methodology also was used to study the influence of temperature on spore germination. The moisture content of the wood blocks was adjusted to above the FSP because experiments showed that germination frequency was independent of the wood moisture content at levels above FSP. The "spore sandwiches" were inserted between the sterile blocks which then were suspended in cheesecloth bags over distilled water in tightly stoppered 250 ml Erlenmeyer flasks. The block cultures were incubated at four different temperatures for 4 days, then the "spore sandwiches" were examined for spore germination. After removal of the "spore sandwiches", the blocks were weighed and their moisture contents were calculated. Three replicate experiments of 16 blocks (four blocks per temperature) were conducted.

RESULTS AND DISCUSSION

Ability of Poria carbonica in soil particles to initiate wood colonization

Soil particles represent a large portion of the air-borne particulate matter in the atmosphere and wind is a major factor in suspending soil in the air (22,23). However, man's activities can have a large influence on a local scale. The size of soil particles transported by wind varies from clay particles less than 1 μm in diameter

to soil aggregates with diameters greater than 100 μm (22,23). Particles less than 20 μm in diameter can be transported hundreds of miles as wind-borne aerosols (22), while sand-sized particles are carried shorter distances. The distance traveled is a function of particle size, wind speed, and turbulence (22).

The smallest soil particles tested for initiation of wood colonization (38 μm) may carry viable P. carbonica propagules capable of establishing colonies in wood (Table 1). Conidia and hyphae in the soil probably were killed during drying. Thus, chlamydospores or particles composed of soil and one or more chlamydospores (9-16.5 X 7-12 μm) (63) initiated wood colonization. The soil may act as a buffer from environmental extremes, allowing longer survival of the chlamydospores. P. carbonica could grow from decayed wood debris into the surrounding soil where, during dry conditions, soil particles similar to those used in these experiments could become air-borne by wind erosion, or movement of heavy machinery.

The influence of light on the production of basidiospores

To investigate the influence of light on sporophore and basidiospore production by P. carbonica, five different isolates were used. Thirty petri plates of malt medium II with Douglas-fir heartwood blocks were inoculated with mycelium of each isolate, and the cultures were incubated at 30°C until colony diameter was about 2

Table 1

COLONIZATION OF DOUGLAS-FIR HEARTWOOD BLOCKS BY *PORIA CARBONICA* ASSOCIATED WITH SOIL PARTICLES OF VARIOUS SIZES

Sieve opening (μm)	Percent blocks colonized[a]
990	100
250	88
175	86
105	97
61	86
38	97

a. Data are based on three replicates of 12 blocks per particle size, incubated at 22°C, 100% RH. The percent colonized blocks was determined by isolating the decay fungus from the wood blocks. There were no significant differences between the percent blocks colonized by different particle sizes (see appendix for statistics).

cm. The cultures were then inverted, one half under diffuse light conditions and the other half were enclosed in foil or 4 mil black plastic to exclude light. After incubating 1 month at about 22°C, the cultures were examined for sporophores and basidiospore production.

Most of the light-incubated cultures produced sporophores, and some cultures of each isolate shed basidiospores onto the plate covers (Table 2). None of the dark-incubated cultures produced any sporophores or spores.

The possibility that restricted gas-diffusion limited sporophore production in the foil- and black-plastic-enclosed cultures was tested in a preliminary experiment. Cultures wrapped in clear plastic were incubated with unwrapped cultures and foil-wrapped cultures. The unwrapped and plastic-wrapped cultures produced sporophores and basidiospores, while the foil-wrapped cultures remained undifferentiated. These results strongly suggest that light is required for P. carbonica sporophore and basidiospore production.

Ability of single chlamydospores to initiate wood colonization

To determine if single chlamydospores were capable of establishing P. carbonica in wood, 25 µl of a chlamydospore suspension was placed on water agar and malt medium I in petri plates, and spread with a glass rod. Single

Table 2

EFFECT OF LIGHT ON SPOROPHORE AND BASIDIOSPORE PRODUCTION BY PORIA CARBONICA IN CULTURE

	Light treatment[b]	Number of cultures of five P. carbonica isolates with sporophores and/ or basidiospores[a]				
		2G	7A	3K	64	LS
Sporophores present	light	22	22	22	15	43
	dark	0	0	0	0	0
basidiospores present	light	6	15	12	7	24
	dark	0	0	0	0	0
Total no. of cultures	light	22	22	22	21	43
	dark	22	22	22	22	37

a. Cultures were incubated at 30°C until the colony diameter reached about 2 to 3 cm then inverted at 22°C in either the light or dark. Total incubation time was about 1 month

b. Cultures in dark were enclosed in either 4 mil black plastic or aluminum foil to exclude light.

spores were picked from the water-agar plates and placed on the endgrain of Douglas-fir heartwood blocks (2 X 2 X 1 cm) with moisture contents above the FSP. Spores on the malt agar were examined microscopically after 3 days and germination percentages were calculated by examining at least 400 spores. After incubating the blocks for 1 month at 22°C, block colonization was determined (general methods).

Single chlamydospores were capable of establishing P. carbonica colonies in Douglas-fir under ideal conditions (Table 3), with about the same level of success as germination on malt medium. Although the probability of a single P. carbonica chlamydospore landing on a pole is low, colony establishment may occur in locations with high P. carbonica inoculum levels.

Moisture content of the "spore sandwiches"

To cause significant decay, basidiomycetes require wood at or above the fiber saturation point (76), but the influence of wood moisture content on spore germination and the initial colonization of wood has not been studied critically. Most basidiospores require relative humidities (RH) above 99% to germinate (79), and regulation of RH is usually achieved by incubating spores over saturated salt solutions (55,58,102,106). Spore germination on thin wood-sections has been reported at 89%

Table 3

GERMINATION OF CHLAMYDOSPORES AND COLONIZATION OF DOUGLAS-FIR HEARTWOOD BLOCKS BY SINGLE *PORIA CARBONICA* CHLAMYDOSPORES

Replicate[a]	Germination percent[b]	Colonization percent[c]
I	60	50
II	78	72

a. Each replicate consisted of 32 blocks and one malt agar plate for spore germination.
b. Percent germination was calculated from examination of 400 chlamydospores on malt extract agar.
c. Percent colonization was calculated from four isolations from the interior of each block.

RH (106), although a free film of water is generally thought necessary. A recent study reported basidiospore germination at humidities as low as 96% RH on thin wood-sections (58), but at relative humidities above 90%, a change of 1°C can cause saturation of the air and condensation of water. Thus, without carefully regulated temperature control, the results of these experiments are questionable.

To compare the moisture content of the two sections comprising the "spore sandwich" with that of the blocks, blocks were prepared in the standard manner and placed over several different salt-solutions or distilled water that produced wood moisture contents ranging from 10 to 86% (based on oven-dry weight). "Spore sandwiches" were wetted with 25 µl of distilled water to simulate inoculation with a spore suspension and placed between the blocks. After 4 days, the sections were removed, quickly weighed, oven dried (24 hr at 110°C), and reweighed. The blocks were also weighed and moisture contents were calculated for the blocks and the sections they contained.

Moisture contents of the wood sections were generally lower than the moisture contents of the blocks surrounding them (Fig. 1). The regression line does not fit the data particularly well ($r^2 = 0.55$), which indicates considerable unaccounted for variation. This variation was mostly due to rapid drying of the thin wood sections before and

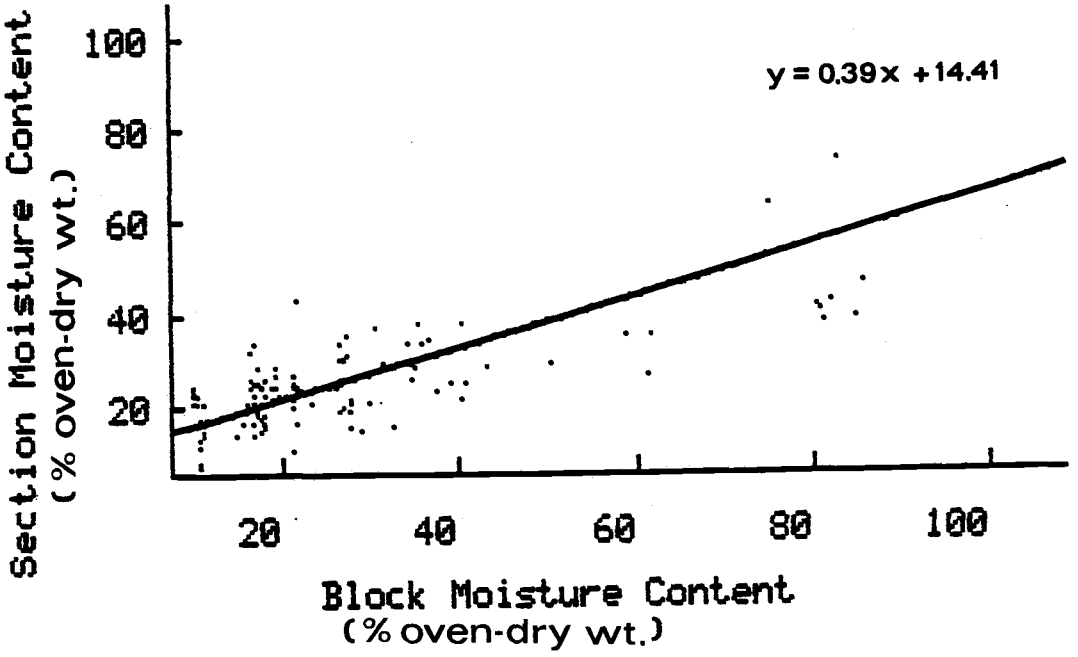


Fig. 1. Moisture content of Douglas-fir heartwood blocks (2 X 1 X 1 cm) used to enclose the "spore sandwiches" compared to the moisture content of the thin wood sections (8 X 8 mm X 60 μm) comprising the "spore sandwiches" after 4 days incubation at 22°C . Results based on 112 blocks.

during weighing; the "spore sandwiches" probably were at the same moisture content as the blocks which enclosed them during incubation.

Influence of wood moisture content on spore germination and wood colonization

Poria carbonica chlamydospores and basidiospores failed to germinate or colonize Douglas-fir heartwood blocks below the FSP, but could germinate and colonize wood at or above the FSP (Table 4). There was no significant difference between basidiospore germination in blocks at and above the FSP; however, chlamydospore germination was significantly lower in blocks at fiber saturation (see appendix for statistics). In an additional experiment with blocks at around 24% or 30% moisture content (MC), chlamydospores also failed to germinate on wood at the lower MC.

These results suggest that poles may be colonized by P. carbonica when their MC exceeds the FSP. Moisture contents in this range commonly occur in freshly cut poles, at the soil-pole contact zone in poles in service, and in checks in poles where rainwater may be trapped. Thus, wood in these conditions is likely to be colonized by decay fungi unless protected.

Table 4

EFFECT OF WOOD MOISTURE CONTENT ON *PORIA CARBONICA* SPORE GERMINATION AND COLONIZATION OF DOUGLAS-FIR HEARTWOOD

Spore type and wood moisture content (%)[a]		Germination percent[b]	Colonization percent[c]
Chlamydospores			
17	(15-18)	0	0
29	(25-35)	71	100
60	(38-81)	90	100
Basidiospores[d]			
15	(15-16)	0	0
35	(31-39)	92	100
83	(71-99)	90	100

a. Data represents three replicate tests of 18 blocks each. Average percent wood moisture content with the range in parentheses.
b. Chlamydospore germination for the two higher moisture contents were significantly different, while basidiospore germination was not (see appendix for statistics).
c. Percent colonization is based on the number of successful isolations of *P. carbonica* from blocks incubated for 1 month.
d. The higher moisture content of the blocks above the fiber saturation point used for basidiospore germination was due to condensation on the blocks caused by a faulty temperature controller.

Influence of temperature on spore germination and wood colonization

The influence of temperature on basidiospore germination of wood decay fungi varies significantly among species. For example, Polyporous tomentosus basidiospores germinated best between 15-27°C (99), while those of Fomes igniarius germinated between 20 and 35°C, but above 30°C the germ tubes were generally short and stunted (23). Pleurotus ostreatus basidiospores germinated between 1 and 30°C, but required 8 days at 1°C and 3 days at 30°C (54). Most of these studies were done using malt extract medium, but because of the nutrient differences, spore germination on wood could be significantly different.

The temperature range for Poria carbonica basidiospore germination was more limited than that found for chlamydospores (Table 5). Basidiospores failed to germinate at 5 and 35°C, although some spore swelling was observed at 35°C. Their germination was significantly lower at 30°C than at 22°C (see appendix for statistics), but wood colonization occurred at both temperatures. Many of the ungerminated basidiospores at 30°C were swollen after 4 days incubation suggesting that they might germinate later.

SEM examination of wood sections from "spore sandwiches" incubated at 22°C showed that P. carbonica basidiospores started to swell after 48 hr, and germinated

Table 5

EFFECT OF TEMPERATURE ON *PORIA CARBONICA* SPORE GERMINATION AND COLONIZATION OF DOUGLAS-FIR HEARTWOOD

Temp. (°C)	Wood moisture content (%)[a]		Germination percent[b]	Colonization percent[c]
Chlamydospores				
5	106	(95-137)	20	0
22	112	(73-151)	90	100
30	89	(71-113)	86	100
35	116	(94-157)	41	0
Basidiospores				
5	93	(64-117)	0	0
22	93	(67-115)	95	100
30	94	(80-106)	33.5	100
35	91	(72-112)	0	0

a. Average percent moisture content with the range in parentheses. Data represents three replicate tests of 16 blocks each.
b. Percent basidiospore germination at 22°C was significantly different than at 30°C. Chlamydospore germination at 5 or 35°C was significantly different than at 22 or 30°C (see appendix for statistics).
c. Percent colonization was calculated from the results of two isolations from each block after a 1 month incubation.

during the next 24 hr period (Fig. 2). Four days after inoculation, the germ tubes were greater than eight spore diameters and wood colonization was beginning.

All Poria carbonica isolates from the blocks colonized by basidiospore inoculum were examined microscopically and were found to be dikaryotic. Because of the large numbers of spores on the wood, hyphal fusion and nuclear exchange between compatible monokaryotic hyphae produced by the germinated basidiospores was guaranteed.

P. carbonica chlamydospores germinated readily and colonized Douglas-fir heartwood blocks at 22 or 30°C (Table 5), but germination frequencies at 5 or 35°C were significantly lower than those at 22 or 30°C (see appendix for statistics). After 3 days incubation at 22 or 30°C, the chlamydospore germ-tube lengths were between 100 and 1000 µm, while those at 5 or 35 C ranged between 10 and 100 µm, but were usually less than 50 µm. P. carbonica chlamydospores incubated at 5 or 35°C were unable to establish colonies 1 mm into the blocks.

The germinating spores incubated at 5°C showed limited germination and colonization probably due to slowed fungal metabolism (24). These spores probably were not killed, and might have resumed growth had the temperature been raised.

Temperatures between 22 and 30°C were favorable for wood colonization by P. carbonica chlamydospores and

Fig. 2. Scanning electron micrographs of germinating Poria carbonica basidiospores on Douglas-fir heartwood incubated at 22°C, (X2000). (a,b) basidiospores swelling after 48 hr incubation; (c,d) germ tubes emerging after 72 hr incubation; (e) germ tubes greater than 5X spore length after 96 hr incubation; and (f) hyphae and ungerminated basidiospore (arrow) after 96 hr incubation.

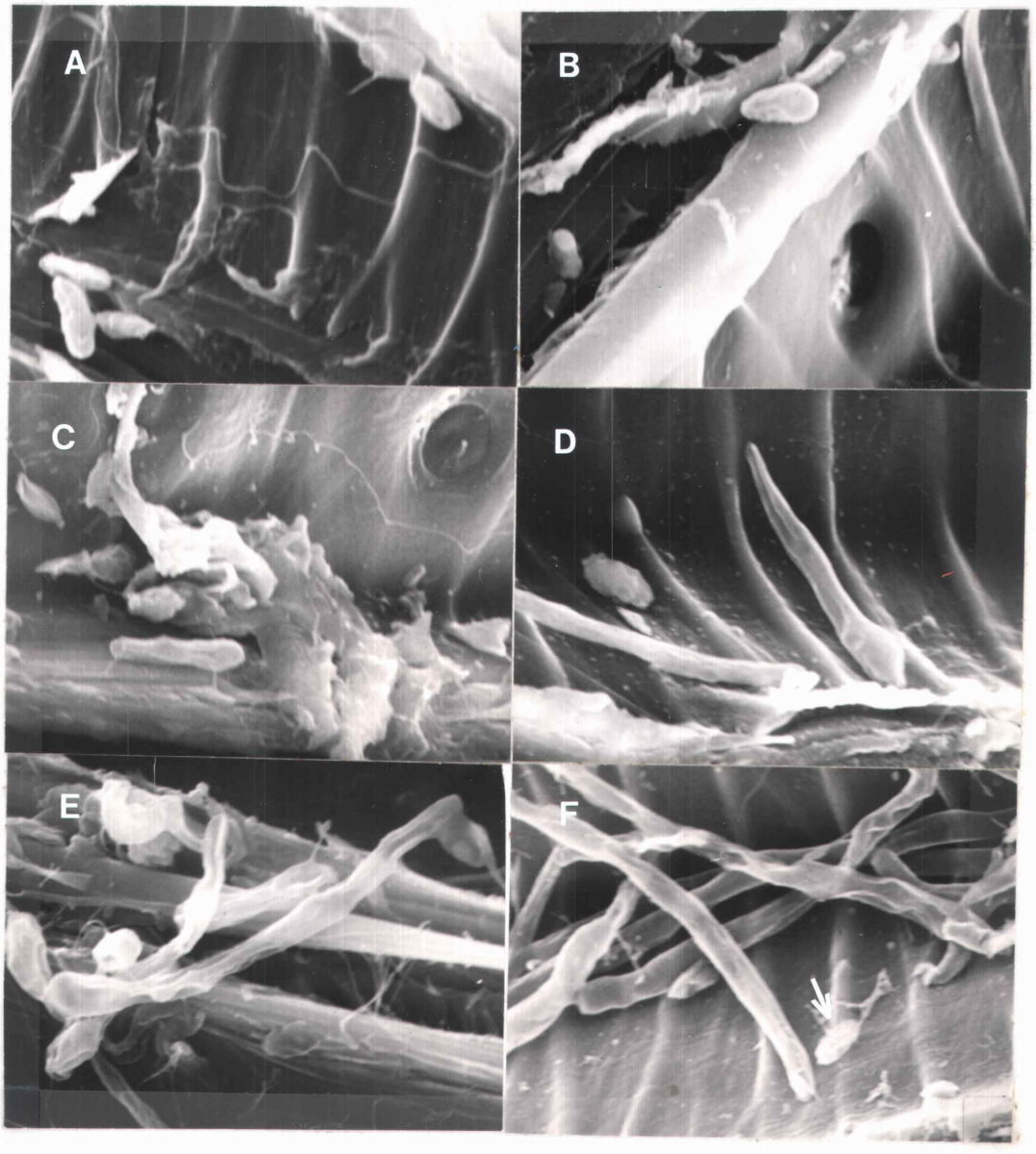


Fig. 2.

basidiospores. These values are comparable to those reported for other wood-decaying basidiomycetes (10,23,24,54,99). Successful colonization of Douglas-fir poles probably would decrease rapidly during periods when temperatures are outside of this range, although spores may survive until more favorable conditions occurred. Temperatures between 5 and 22°C need to be tested to establish the minimum colonization temperature.

Ability of basidiomycete monokaryons to decay wood

Basidiospores are a major means of dissemination of basidiomycetes, therefore the monokaryotic hyphae arising from single spores should be capable of colonizing and decaying wood. Investigations comparing the ability of monokaryons and dikaryons to cause decay showed that dikaryons of Lenzites trabea were more destructive than the monokaryons (1,13). However, Poria vaillantii (13) and Serpula lachrymans (15) monokaryons generally were more destructive than dikaryons.

Poria placenta and P. carbonica are closely related members of the same genus and produce a similar decay in wood. They are often isolated together from decaying Douglas-fir poles (104), and produce similar appearing colonies in culture, although P. placenta grows faster and lacks perpendicular-branching skeletal hyphae. Samples taken from air-seasoning poles showed that these two fungi

have similar patterns of distribution within the poles (Chap. 3), and probably respond similarly to environmental factors.

The abilities of mono- and dikaryons of P. placenta to cause decay were compared using an agar block test (5). French square bottles were filled with 20 ml of malt extract medium I and laid on their sides while the agar solidified. After cooling, the medium was inoculated with four dikaryotic or four monokaryotic isolates of P. placenta, seven bottles per isolate (isolates obtained from the Center for Forest Mycology Research, U.S. Forest Products Laboratory, Madison, WI). The cultures were incubated at 30°C until a mycelial mat covered the medium surface. Douglas-fir heartwood blocks then were placed on sterile glass supports on the medium surface, two blocks per chamber. The blocks had been oven dried (110°C for 24 hr), weighed, autoclaved and vacuum saturated with sterile distilled water prior to placement in the bottles. After 4 months incubation at 30°C, the blocks were removed, scraped free of mycelium, oven dried, and reweighed. The monokaryotic isolates were checked for the presence of clamps, and none were found.

There were no significant differences between the weight losses caused by monokaryons and dikaryons (Table 6) (see appendix for statistics). There were significant differences between the isolates in each group, which were

Table 6

WEIGHT LOSS OF DOUGLAS-FIR HEARTWOOD BLOCKS INOCULATED WITH DIKARYOTIC OR MONOKARYOTIC ISOLATES OF *PORIA PLACENTA*

Isolate	Percent weight loss after 4 months incubation at 30°C[a]
Monokaryon	37.5
Dikaryon	36.5
Dikaryon	33.9
Dikaryon	32.5
Dikaryon	20.1
Monokaryon	19.9
Monokaryon	15.2
Monokaryon	13.9
Average of Dikaryons	30.7
Average of Monokaryons	21.7

a. Weight loss based on oven-dry weight. Values represent means of 14 blocks per isolate. There was no significant difference between the dikaryons and the monokaryons (see appendix for statistics).

greater than the differences between the groups. Generally, the monokaryons caused less weight loss than the dikaryons, but a monokaryon caused the greatest weight loss.

These results demonstrated that monokaryotic isolates of P. placenta, arising from single basidiospores, were capable of establishing themselves and causing serious decay without fusing with a compatible monokaryon to form a dikaryon. The large variation in decay capability between isolates of the same species emphasizes the danger of relying on one isolate for laboratory tests.

General discussion

Results of these experiments indicate that temperature and wood moisture content are important factors that define favorable periods for wood colonization by decay fungi. While temperature seldom would be limiting in the Pacific Northwest's mild maritime climate, optimum moisture levels favorable for wood colonization by basidiomycetes are more likely to occur during the wet winter months.

Because the outer surface of utility poles contains preservatives, spores landing on this surface are less likely to be successful than those that fall into seasoning checks. For example, a single chlamydospore-containing soil particle landing on untreated wood exposed

by checking could initiate a P. carbonica colony in a pole when moisture and temperatures were favorable.

The "spore sandwich" methodology developed herein places spores in an environment surrounded by wood at a specific temperature and moisture content. The airflow around the "spore sandwich" was restricted, which reduced the wetting and drying that occurs in poles in service, nevertheless the "spore sandwich" created an environment similar to that surrounding spores in seasoning checks where successful colonization generally takes place (10,16,27,34,104).

Spore germination is the stage in the decay fungus life cycle most susceptible to inhibitory chemicals, because at this time the fungus is poorly established in the wood, and has few food reserves (84). Preservatives and fumigants that remain effective for many years probably prevent spore germination by decay fungi. Using the "spore sandwich" method it is possible to evaluate new fungicides against P. carbonica basidiospores on wood in the laboratory. The results obtained should correlate more closely with those obtained in the field, thus enhancing the development of effective treatments that ensure long service life for preservative-treated wood.

CHAPTER 2

Environmental Influences on the Colonization of Douglas-fir Pole Sections by Basidiomycetes

INTRODUCTION AND LITERATURE REVIEW

To determine when air-seasoning poles were being colonized and by what fungi, estimates of the basidiomycete inoculum levels in the yards throughout the year were needed. Records of seasonal variations in temperature and moisture, which affect spore production, release, germination, and growth also were needed. Correlation of these environmental factors with inoculum levels should allow identification of periods with high potential for decay initiation. This knowledge could be used to modify seasoning practices to minimize decay.

Detection of wood-decay fungi spores in the environment

Unlike many plant pathogens that produce extremely large numbers of distinctive spores, basidiospores of different wood-decay fungi appear similar and usually occur in much smaller numbers (101). This limits the usefulness of standard spore trapping devices which collect spores to estimate the potential for a plant disease to occur in epidemic proportions.

Wood discs have been used as spore traps to study the seasonal variation in spore deposition and colonization of wood by basidiomycetes (44,71,72,89). Only those spores capable of germinating and growing on wood were detected.

The distribution of wood-decaying basidiomycetes in Finland (44) was studied using discs of freshly cut spruce exposed for 2 or 4 hours. The discs were incubated for 2 weeks following exposure, and then the basidiomycetes were isolated and identified.

Seasonal variation in spore deposition by Heterobasidion annosum in British Columbia forests was examined using discs of freshly cut, white pine (71) which were exposed for 2 hours every 2 weeks. Spore populations were highest in October and November when the temperature was between 0 and 6°C.

A study of the seasonal variation in infection of Douglas-fir logs and stumps by H. annosum (72) used 24 to 32 foot Douglas-fir logs (12 inch diameter). Several trees were felled each month for 1 year, left for 2 years in the woods, and then sampled. Log and stump colonization was highest in logs felled from March to August.

Another technique for detecting specific basidiomycetes used actively growing monokaryotic cultures of the fungus sought (101). The cultures were exposed for specific time periods, then incubated under conditions conducive to spore germination, and examined microscopically to detect dikaryotic mycelium. Basidiospores, that germinated on the medium, produced monokaryotic hyphae which fused with the surrounding hyphae to produce

dikaryotic mycelium. The number of dikaryons gave an estimate of the viable basidiospores in the environment.

The use of culture plates of monokaryotic basidiomycetes as spore traps was unsuitable for this study because spores of only one basidiomycete would be detected. Wood discs, which dry rapidly and require frequent replacement, were not practical due to the distances between study sites and the duration of the study. Instead, small pole sections exposed for at least 3 months were used as spore traps.

Objectives

The objectives of this study were: (i) to estimate the colonization frequency of Douglas-fir pole sections by basidiomycetes at four Pacific Northwest air-seasoning yards throughout the year; (ii) to identify the basidiomycetes isolated from the pole sections; (iii) to compare colonization frequency of the endgrain with that of the longitudinal surfaces of the pole sections; and (iv) to examine temperature and precipitation records as predictive factors for periods of high potential for wood colonization by basidiomycetes.

MATERIALS AND METHODS

Sterilized pole sections were exposed for 3-month periods at four widely separated sites in the Pacific Northwest. After exposure, the sections were sampled and

the decay fungi present in the samples were cultured and identified.

Preparation of the pole sections

Douglas-fir pole sections (8.5 ft long) were coated on both ends with three coats of Gaco A5400 paint to retard moisture loss, prior to steam sterilization in a kiln for 48 hr at 100°C. Initially, pole sections from each yard were sampled prior to exposure to ensure they were free of decay fungi. Unfortunately, many sections contained basidiomycetes; thus, new colonies could not be distinguished from expansion of existing colonies. To avoid this problem, all subsequent pole sections were sterilized prior to exposure.

Pole section exposure at four sites in the Pacific Northwest

Four widely separated locations were chosen in the Pacific Northwest to establish the experimental plots: Arlington, WA; Scappoose, OR; Eugene, OR; and Oroville, CA.

Four pole sections (8.5 ft long, 12 in diameter) were transported to each exposure site where about twelve 6-inch cores were removed around the circumference 2 feet in from each end to verify sterilization. Only six of 833 cores taken from sterilized pole sections during this study contained possible basidiomycetes. The only isolate identified was a monokaryon of Phlebia "A", a basidio-

mycete commonly found on slash and wood debris, that probably colonized the pole section shortly after sterilization.

Following core removal, a 2-foot segment was cut from both ends of each pole section, adjacent to the coring holes, and the wood around the coring holes was removed. This divided the original pole section into two 2-foot segments, each with one coated end, and a 4-foot segment with uncoated ends. Eight 2-foot and four 4-foot segments were placed at each plot location. The 2-foot sections were set vertically on the coated ends to expose the freshly cut cross-section, while the 4-foot sections were laid horizontally on treated skids. Eight endgrain faces were exposed horizontally and eight faces were exposed vertically. The pole sections were exposed at each location for either 3 or 6 months after which they were replaced with a new set of sections over a 21-month period.

Initially, a 6 month exposure period was included because it was not known if 3 months was sufficient time for basidiomycetes to become established in the wood in detectable numbers. Relatively large numbers of basidiomycetes were isolated from sections exposed for 3 months so the 6 month exposures were discontinued after the first year.

Sampling of the pole sections

The pole sections were returned to the Forest Research Laboratory where moisture contents were measured immediately using a resistance-type moisture meter. Readings were taken at depths of 0.5, 1.0 and 2.0 inches. The 2-foot sections were measured at the ends, 6 and 12 inches in from each end, and in the middle, on two sides. Moisture readings were taken on the 4-foot sections at both ends, 9 and 18 inches in from each end, and in the middle, on the upper and lower surfaces.

About 24 cores were removed around the circumference 1 and 12 inches from the upper end of each 2-foot section (vertical exposure). Each 4-foot section (horizontal exposure) was cored around the circumference 1 inch from each end, and at 4 inch intervals in two parallel rows along the upper surface, offset 3 inches from the center, for a total of about 34 cores. Following removal, the cores were cultured for basidiomycetes as described in the general methods section. The unidentified fungi were separated into basidiomycetes and suspect fungi which had basidiomycetous characteristics, but lacked clamp connections. The terms "basidiomycetes" and "decay fungi" in this chapter refer to all the basidiomycetes and suspect fungi isolated from the poles.

Location of weather stations

Weather data for this study were obtained from the U.S. Climatological Service weather station nearest to each plot. Data from a Seattle, WA (urban site) weather station was used for Arlington, WA. Arlington is about 45 miles north of Seattle, 20 miles east of Puget Sound, and experiences the same maritime climate as Seattle. Data from the Portland airport which is about 30 miles from the Scappoose, OR, site was used. The data for Eugene, OR was obtained at the Eugene airport, which is about 4 miles from the study site, and data from Red Bluff, CA (60 miles from the study site in Oroville) was used since both are located on the floor of the Sacramento Valley, and experience similar weather patterns.

RESULTS AND DISCUSSION

Effect of sterilization on wood colonization by basidiomycetes

Autoclaving or steaming of wood blocks subsequently increased basidiospore germination on wood (59,82), the basidiomycete growth rate in the wood, and the wood weight loss from decay (11,77,78,86). These increases probably were due to concurrent changes that occur in wood heated to 135^{o}C for 2 or more days, including changes in the lignin and cellulose structures (33), and an increase in soluble compounds (11,77,78,86). Douglas-fir lost about 7% of its decay resistance after sterilization (78).

To determine the effects of kiln sterilization of pole sections on colonization by decay fungi, ten pole sections (10 ft long) were sampled by removing 6-inch cores to determine the initial basidiomycete frequency in the wood and then a 5 foot segment was cut from each section and steam-sterilized in a kiln at 100°C for 48 hours. The sterile and non-sterile pole sections were exposed for 1 year at the Northwest Forest Genetics Center, Corvallis, OR.

Initially, 21% of 660 cores from the pole sections contained basidiomycetes. After 1 year, 41% of 936 cores from the non-sterile sections and 18% of 936 from the sterile pole sections contained basidiomycetes. Although appearing to be significantly different, the results were inconclusive because establishment of new basidiomycete colonies in the non-sterile sections could not be distinguished from expansion of colonies initially present in the wood.

Sterilization of pole sections prior to exposure may have increased the soluble carbohydrates in the wood which might have affected the amount and the species composition of fungi that colonized the pole sections. Possible effects include enhanced spore germination and growth in wood, and increased colonization of the pole sections by competing fungi. Nevertheless, sterilization of the pole sections was necessary to detect newly established

basidiomycete colonies in the wood.

Frequency of basidiomycetes isolated from sterilized pole sections exposed for 3 month intervals at four sites in the Pacific Northwest

Colonization occurred throughout the year at all four locations (Table 7). With the exception of the period from November, 1981 to January, 1982, the basidiomycete isolation frequency was low (0 to 10%). Isolation frequency increased significantly in the period from November, 1981 to January, 1982 in three of the four sites. During this period, basidiomycete isolation frequency increased from north to south. Arlington, WA (the northern most location) showed no increase during the time period from November, 1981 to January, 1982, but had a significant increase during May through July, 1982.

Influence of precipitation on colonization of Douglas-fir pole sections

Adequate moisture is necessary for basidiospore release, germination, and growth in wood. Interestingly, at three of the four plot locations, the number of days with measurable precipitation was significantly higher during the period from November, 1981 to January, 1982 than during any other 3 month period (Table 8). The higher basidiomycete isolation frequency from pole sections exposed during this period may have been partially due to the increased precipitation. Higher fungal inoculum levels i.e. more basidiospores, increased

Table 7

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT FOUR SITES IN THE PACIFIC NORTHWEST

Plot Location	Begin / End	Percentage of cores containing basidiomycetes removed from pole sections exposed for three month intervals.[a]						
		5/81 7/81[b]	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Arlington, WA		3.7	5.9	4.6	1.8	9.7	3.2	1.2
Scappoose, OR		5.1	6.6	15.3	5.2	3.1	1.1	1.2
Eugene, OR		-[c]	8.1	31.9	2.1	0.7	1.9	1.7
Oroville, CA		3.8	5.0	34.2	5.9	1.5	3.1	0

a. Results based on an average of 270 cores (range 183-360) removed from 12 pole sections at each plot location during each exposure period.
b. Pole sections in the first exposure period were not sterilized.
c. Results lost due to contamination of the culture plates by a Neurospora sp.

Table 8

NUMBER OF DAYS WITH DETECTABLE PRECIPITATION
AT FOUR LOCATIONS IN THE PACIFIC NORTHWEST

Weather Station Location[a]	Number of days with detectable precipitation during 3 month intervals. Begin / End	5/81 7/81	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Seattle, WA		49	39	72	54	39	46	53
Portland, OR		46	32	74	64	26	44	54
Eugene, OR		20	39	71	50	30	40	57
Red Bluff, CA		11	14	52	35	9	22	52

a. Weather data was from the U.S. Climatological Service weather station closest to each plot location.

wood colonization success of propagules, or more basidiomycete growth in the pole sections could have caused the higher frequencies.

Influence of temperature on colonization of Douglas-fir pole sections

The number of days with temperatures conducive to fungal growth may have influenced basidiomycete colonization of the pole sections. The number of days with temperatures greater than 10°C was higher during the period from November, 1981 to January, 1982 than during the same period the following year (Table 9). Higher temperatures would increase fungal growth rates, resulting in larger colonies in the wood, thereby increasing the isolation frequency of the decay fungi.

The number of days with precipitation at Red Bluff was the same during the periods from November, 1981 to January, 1982, and November, 1982, to January, 1983 (ie. 52 days). However, the increased basidiomycete isolation frequency from pole sections exposed during the earlier period could have been due to the higher temperatures during this period.

The number of days with temperatures above 10°C was lowest in Seattle during the period from November, 1981 to January, 1982, and this may explain the failure of basidiomycetes to colonize the pole sections exposed in Arlington when moisture levels were favorable for

Table 9

NUMBER OF DAYS WITH AN AVERAGE DAILY TEMPERATURE EQUAL TO OR GREATER THAN 10 C. AT FOUR LOCATIONS IN THE PACIFIC NORTHWEST

Weather Station Location[a]	Number of days with an average temperature above 10°C during 3 month intervals. Begin 5/81 End 7/81	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Seattle, WA	93	94	32	60	92	94	24
Portland, OR	92	96	47	93	73	90	36
Eugene, OR	91	87	35	78	99	76	17
Red Bluff, CA	92	93	81	85	99	87	69

a. Weather data was from the U.S. Climatological Service weather station closest to each plot location.

colonization. Eugene had fewer warm days than Portland during this period, yet the basidiomycete isolation frequency was higher from sections exposed in Eugene than sections exposed in Scappoose. This may have resulted from a higher local basidiospore inoculum level at the Eugene plot. Several log-storage yards with large volumes of wood and debris that may have contained decay fungi were adjacent to the plot.

As discussed earlier, spore germination and colony establishment by basidiospores requires wood with a moisture content at or above the FSP, but not so high as to exclude oxygen (79,106). Temperature and precipitation interact to affect wood moisture content (e.g. high temperatures increase the drying rate, and more precipitation is required to keep wood above the FSP, while less precipitation is required at low temperatures). Temperature and precipitation interactions probably increased the isolation frequency of basidiomycetes during the November, 1981 to January, 1982 period. Pole section moisture contents were well above the FSP at all plot locations during this period (Table 10). During the other periods studied, temperature or precipitation were unfavorable and pole section moisture content fell below the FSP; thus, limiting spore germination and wood colonization.

Table 10

MOISTURE CONTENT OF STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT FOUR LOCATIONS IN THE PACIFIC NORTHWEST

Exposure Site	Wood moisture content (% dry wt.) of pole sections exposed for 3 month intervals[a]						
Begin / End	5/81 7/81[b]	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Arlington, WA	15	28	42	20	16	17	27
Scappoose, OR	14	25	41	18	19	20	31
Eugene, OR	15	26	40	16	13	15	29
Oroville, CA	8	23	34	10	8	12	21

a. Measured 2 inches in from the surface of pole sections at the end of the exposure period.
b. Poles sections used in the first exposure period were not sterilized.

The influences of the number of days with temperatures above 10°C (DTEMP), the number of days with detectable precipitation (DPPT), and pole section moisture content (PMC) on the percent colonization of the sterilized pole sections were tested with multiple linear regression. PMC was the best predictor of percent colonization ($p=0.02$), but accounted for only 13% of the variation ($r^2=0.1354$). Addition of DPPT and DTEMP to the model accounted for another 5% of the total variation. However, t-tests of these additional variables showed that DPPT and DTEMP were not significant in this model ($p=0.36$, $p=0.37$ respectively). DPPT and PMC; and DTEMP and PMC were correlated ($r^2=0.55$, $r^2=0.47$ respectively). Thus, DPPT and DTEMP were not significant in this model because they accounted for the same portion of the variation in percent colonization as did PMC. The low overall correlation of the model suggests that one or more major factors that were not directly measured, e.g. inoculum density, were responsible for most of the variation in wood colonization between the four sites.

Basidiomycete species isolated from sterilized pole sections exposed for 3 month intervals

About 30 basidiomycete species were identified from the isolates cultured from sterilized pole sections exposed at the four plot locations (Tables 11-14). Species differed between plot locations and time of

Table 11

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT ARLINGTON, WA

	Number of cores containing basidiomycetes						
	Exposure periods (months)						
Fungus species	Begin 5/81 End 7/81[a]	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Poria placenta (dikaryon)	1	3	0	0	5	0	0
monokaryon	0	0	2	0	2	2	0
Stereum hirsutum	0	0	2	0	0	0	0
Peniophora spp.	0	2	3	0	0	0	0
Sistotrema brinkmanii	0	0	1	0	0	0	0
Phanerochaete sordida	1	1	0	0	0	4	0
Poria xantha (dikaryon)	0	0	0	0	0	1	0
monokaryon	0	0	0	0	1	0	0
Epicoccum nigrum[b]	0	0	0	0	1	0	0
Coriolus versicolor (monokaryon)	0	0	1	0	0	1	1
Phlebia radiata (dikaryon)	0	1	0	0	0	0	0
monokaryon	1	0	0	0	0	0	0
Phlebia 'A' (monokaryon)	0	0	0	0	0	1	0
Poria carbonica (dikaryon)	0	1	0	0	1	0	0
monokaryon	0	1	0	0	0	0	0
Phlebia gigantea	0	0	0	0	0	1	0
Poria cinerascens (monokaryon)	0	0	0	0	0	0	1
Phlebia albida	0	0	1	0	0	0	0
Unidentified basidiomycetes	4	2	1	2	2	0	0
Unidentified suspect fungi[c]	2	1	4	3	16	1	0
Number of cores with fungi[d]	8	12	14	5	26	10	3
Total cores taken	218	204	303	286	267	317	253
Percent cores with fungi	3.7	5.9	4.6	1.8	9.7	3.2	1.2

a. Pole sections in the first exposure period were not sterilized.
b. Not a basidiomycete.
c. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
d. Does not equal the sum of the column as cores may have more than one fungus.

Table 12

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT SCAPPOOSE, OR

Number of cores containing basidiomycetes

	Exposure periods (months)						
Fungus species / Begin / End	5/81 7/81[a]	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Poria placenta (dikaryon)	0	0	3	2	0	0	0
monokaryon	1	5	8	1	1	1	0
Stereum hirsutum	3	1	1	0	1	0	0
Peniophora spp.	0	1	18	0	0	0	0
Sistotrema brinkmanii	0	1	1	0	0	0	0
Phanerochaete sordida	0	3	0	0	3	2	0
Haematostereum sanguinolentum	4	0	1	3	0	0	0
Poria xantha (monokaryon)	0	0	4	0	0	0	0
Coriolus versicolor (dikaryon)	1	0	0	1	0	0	0
monokaryon	0	0	0	1	1	0	0
Phlebia radiata (dikaryon)	0	0	1	0	0	0	0
monokaryon	0	0	1	0	0	0	0
Phlebia 'A' (monokaryon)	0	0	1	0	0	0	0
Phlebia gigantea	0	0	0	0	0	0	1
Heterobasidion annosum	0	0	0	1	0	0	1
Poria cinerascens (monokaryon)	0	0	0	1	0	0	1
Unidentified basidiomycetes	1	1	3	0	0	0	0
Unidentified suspect fungi[b]	7	2	3	6	2	1	0
Number of cores with fungi[c]	17	14	44	15	8	4	3
Total cores taken	335	211	288	288	257	360	261
Percent cores with fungi	5.1	6.6	15.3	5.2	3.1	1.1	1.2

a. Pole sections in the first exposure period were not sterilized
b. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
c. Does not equal the sum of the column as cores may have more than one fungus.

Table 13

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT EUGENE, OR

	Number of cores containing basidiomycetes					
	Exposure periods (months)					
Fungus species / Begin / End	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Poria placenta (dikaryon)	0	5	1	0	0	0
monokaryon	0	11	1	0	0	2
Stereum hirsutum	0	9	1	0	1	1
Peniophora spp.	0	16	0	0	0	0
Sistotrema brinkmanii	0	30	0	0	0	0
Haematostereum sanguinolentum	0	3	0	0	0	0
Poria xantha (monokaryon)	0	0	0	0	1	0
Epicoccum nigrum[a]	0	0	2	1	0	0
Coriolus versicolor	0	0	0	0	3	0
Unidentified basidiomycetes	11	16	0	0	0	1
Unidentified suspect fungi[b]	6	6	1	1	0	1
Number of cores with fungi[c]	17	92	6	2	5	5
Total cores taken	209	288	280	281	269	293
Percent cores with fungi	8.1	31.9	2.1	0.7	1.9	1.7

a. Not a basidiomycete.
b. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
c. Does not equal the sum of the column as cores may have more than one fungus.

Table 14

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS AT OROVILLE, CA

	Number of cores containing basidiomycetes						
	Exposure periods (months)						
Fungus species	Begin 5/81 End 7/81[a]	8/81 10/81	11/81 1/82	2/82 4/82	5/82 7/82	8/82 10/82	11/82 1/83
Poria placenta (dikaryon)	1	0	12	0	0	0	0
monokaryon	1	0	16	0	0	0	0
Stereum hirsutum	0	1	25	7	0	2	0
Peniophora spp.	0	2	21	1	0	0	0
Sistotrema brinkmanii	0	2	2	0	0	1	0
Phanerochaete sordida	1	0	17	2	0	0	0
Poria xantha	0	0	3	0	0	0	0
Epicoccum nigrum[b]	0	0	1	1	0	0	0
Coriolus versicolor (dikaryon)	1	0	0	1	0	0	0
monokaryon	0	1	0	1	0	0	0
Phlebia radiata (monokaryon)	0	0	1	0	0	1	0
Phlebia 'A' (monokaryon)	0	0	0	0	0	1	0
Poria cinerascens (monokaryon)	0	0	0	0	0	1	0
Fomitopsis cajanderi (monokaryon)	0	1	0	0	0	0	0
Unidentified basidiomycetes	1	1	7	5	0	0	0
Unidentified suspect fungi[c]	2	3	5	6	4	2	0
Number of cores with fungi[d]	7	11	103	17	4	8	0
Total cores taken	183	222	301	288	265	261	275
Percent cores with fungi	3.8	5.0	34.2	5.9	1.5	3.1	0

a. Pole sections in the first exposure period were not sterilized
b. Not a basidiomycete.
c. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
d. Does not equal the sum of the column as cores may have more than one fungus.

exposure, probably due to climactic variations and amounts of local inoculum (e.g. basidiospores).

The frequency of basidiomycete species isolated from the exposed pole sections summed over all 3 month periods increased from north to south (Table 15). The frequency of P. placenta mono- and dikaryons was particularly high, with monokaryons more abundant than dikaryons at three plots.

Poria placenta monokaryons from basidiospores of the same genetic strain will form dikaryons in about 50% of random pairings (62). Dikaryon frequency is higher when monokaryons isolated from different strains are paired randomly because P. placenta has multiple alleles at the mating type locus (62). Thus, the frequency of monokaryotic isolates, relative to the dikaryotic isolates would be higher in locations where only one strain was producing basidiospores, and lower where the basidiospores come from several different strains.

The high frequency of P. placenta monokaryons relative to dikaryons suggests that basidiospores are responsible for a high percentage of the pole colonization by P. placenta, and that basidiospore sources at each location may be relatively homogeneous. The high monokaryon frequency also suggests that 3 months may not be sufficient time for the colonies to grow together and exchange nuclei.

Table 15

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM STERILIZED POLE SECTIONS EXPOSED FOR 3 MONTH INTERVALS OVER A 21-MONTH PERIOD AT FOUR SITES IN THE PACIFIC NORTHWEST

	Number of cores containing basidiomycetes			
	Plot location[a]			
Fungi	Arl.	Sca.	Eug.	Oro.
Poria placenta (dikaryon)	8	5	6	12
monokaryon	6	16	14	16
Stereum hirsutum	2	3	12	35
Peniophora spp.	5	19	16	24
Sistotrema brinkmanii	1	2	30	5
Phanerochaete sordida	10	8	-	19
Haematostereum sanguinolentum	1	4	3	-
Poria xantha (dikaryon)	1	-	-	3
monokaryon	1	4	1	-
Epicoccum nigrum[b]	1	-	3	2
Coriolus versicolor (dikaryon)	-	1	3	1
monokaryon	3	2	-	3
Phlebia radiata (dikaryon)	1	1	-	-
monokaryon	1	1	-	2
Phlebia 'A' (monokaryon)	1	1	-	1
Poria carbonica (dikaryon)	2	-	-	-
monokaryon	1	-	-	-
Phlebia gigantea	1	1	-	-
Heterobasidion annosum	-	2	-	-
Poria cinerascens	-	-	-	1
monokaryon	1	2	-	1
Fomitopsis cajanderi monokaryon	-	-	-	1
Phlebia albida	1	-	-	-
Crustoderma dryinum	-	-	1	-
monokaryon	-	1	-	-
Unidentified basidiomycetes	6	4	26	8
Unidentified suspect fungi[c]	20	13	14	19
Number of cores with decay fungi	70	88	126	143
Total number of cores	1630	1665	1620	1612

a. Arl.= Arlington, WA ; Sca.= Scappoose, OR
Eug.= Eugene, OR ; Oro.= Oroville, CA
b. Not a basidiomycete.
c. Suspect fungi are those isolates which have basidiomycetous characteristics but lack clamp connections.

Frequency of basidiomycetes isolated from pole sections exposed for 6 month intervals

Although pole sections exposed from May, 1981 to November, 1981 were nonsterile before exposure, more species and higher frequencies of basidiomycetes were isolated from the sterilized sections exposed during the following 6-month period (Table 16). This increase was probably due to the absence of competing fungi and decreased decay resistance caused by sterilization. Moreover, the environmental conditions for wood colonization during the second exposure period were favorable (Tables 8-10).

The relatively high isolation frequency of P. placenta monokaryons suggests that basidiospores were prevalent within the plots. P. placenta was more frequent in pole sections exposed at Scappoose and Eugene than in those exposed at Arlington and Oroville. This probably resulted, in part, from higher basidiospore concentrations in Eugene and Scappoose, since there were more potential sporophore-containing logs stored around these yards.

The lower P. placenta isolation frequency at Arlington and Oroville may also be due, in part, to less favorable weather for fungal growth at these plots. Arlington generally experiences cooler temperatures which would slow wood colonization by decay fungi. While Oroville is generally warmer, the dry climate may limit wood

Table 16

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM POLE SECTIONS EXPOSED FOR 6 MONTH INTERVALS AT FOUR LOCATIONS IN THE PACIFIC NORTHWEST

	Percent cores containing basidiomycetes Exposure intervals and location[a]							
	May '81–Nov. '81				Nov. '81–May '82			
Fungi	A	S	E	O[b]	A	S	E	O[c]
Poria placenta (dikaryon)	-	0.8	0.9	-	0.7	26.7	19.3	4.5
monokaryon	0.4	0.8	1.8	-	3.1	13.4	6.8	9.8
Stereum hirsutum	0.4	-	-	0.4	0.7	2.5	3.2	10.1
Peniophora spp.	2.2	-	-	-	1.0	1.1	0.7	2.8
Haematostereum sanguinolentum	6.7	-	-	-	3.1	-	0.4	-
Sistotrema brinkmanii	-	0.4	0.4	-	2.1	-	3.9	-
Crustoderma dryinum	-	-	-	-	0.7	1.1	2.5	-
Poria carbonica (dikaryon)	-	0.8	-	-	-	-	0.4	-
monokaryon	-	1.2	-	-	-	-	-	-
Coriolus versicolor (dikaryon)	-	-	-	-	-	-	0.7	-
monokaryon	-	-	0.4	-	0.3	1.1	-	0.3
Phlebia radiata (dikaryon)	0.9	-	0.4	-	0.3	1.1	-	-
monokaryon	-	-	0.9	-	1.0	-	-	-
Poria xantha (dikaryon)	-	-	-	-	-	2.9	-	-
monokaryon	-	-	-	-	-	0.7	0.4	0.7
Phanerochaete sordida	0.4	0.4	0.4	0.4	0.3	-	-	-
Phlebia 'A' (monokaryon)	-	-	-	-	-	0.3	0.7	-
Unidentified basidiomycetes	1.3	1.6	1.3	0.4	5.6	14.4	7.8	5.9
Unidentified suspect fungi[d]	0.9	3.6	1.3	1.4	2.8	5.8	2.1	10.4
Percent cores with fungi	12.0	12.7	7.6	3.2	20.5	59.6	47.5	41.8
Total cores taken	225	244	224	219	288	277	280	287

a. A= Arlington, WA; S= Scappoose, OR
E= Eugene, OR ; O= Oroville, CA
b. Poles sections were not sterilized before exposure.
c. Pole sections were sterilized before exposure.
d. Suspect fungi are those isolates which have basidiomycetous characteristics but lack clamp connections.

colonization during much of the year.

Colonization of the different pole surfaces

The different pole surfaces sampled on the 2- and 4-foot pole sections were equally susceptible to fungal colonization (Table 17) (see appendix for statistics). However, individual basidiomycete species exhibited distinct distribution patterns, reflecting their relative competitiveness in the different pole zones. For example, P. placenta was isolated more frequently from the the section ends than from the upper pole surfaces which suggests it was colonizing the exposed heartwood. Conversely, Peniophora spp. were isolated more frequently from the upper pole surfaces, probably from the sapwood.

General discussion

As might be expected, the number of species and frequencies of individual basidiomycete species isolated from pole sections exposed for 6 months was much higher than that from sections exposed for 3 months. This was due primarily to the longer incubation time, that allowed colonies in the wood to grow larger, which increased the probability of isolation. For example, Crustoderma dryinum was not isolated from poles sections exposed for 3 months, but was isolated from pole sections exposed for 6 months. However, Peniophora spp. which were abundant in pole sections exposed for 3 months, decreased after 6

Table 17

FREQUENCY AND DISTRIBUTION OF BASIDIOMYCETES ON STERILIZED POLE SECTIONS EXPOSED FOR 3-MONTH INTERVALS AT FOUR LOCATIONS IN THE PACIFIC NORTHWEST

Percent cores containing basidiomycetes

	Vertical 2-foot sections		Horizontal 4-foot sections		
Fungi species	Top	Middle	Top	Upper	Butt
Poria placenta (dikaryon)	0.6	0.5	0.2	0.3	0.6
monokaryon	1.3	0.2	1.3	0.6	0.4
Stereum hirsutum	0.9	0.8	0.6	0.6	1.0
Peniophora spp.	0.4	0.8	0.5	3.4	0.5
Sistotrema brinkmanii	0.5	0.7	0.5	0.6	0.6
Phanerochaete sordida	0.9	0.2	0.7	0.6	0.3
Haematostereum sanguinolentum	0.4	0.1	-	-	-
Crustoderma dryinum	0.1	-	-	-	-
monokaryon	0.1	-	-	-	-
Poria xantha (dikaryon)	0.1	-	-	0.2	0.1
monokaryon	0.1	-	0.2	0.1	0.1
Epicoccum nigrum[a]	-	-	0.3	0.1	0.2
Coriolus versicolor (dikaryon)	0.1	0.1	0.3	0.1	0.1
monokaryon	0.1	0.1	0.3	0.1	-
Phlebia radiata (dikaryon)	-	0.1	-	0.1	-
monokaryon	-	0.1	-	0.1	-
Phlebia 'A' (monokaryon)	-	-	0.1	0.1	0.1
Poria carbonica (dikaryon)	0.1	-	-	0.1	-
monokaryon	0.1	-	-	-	-
Phlebia gigantea	-	0.1	-	-	-
Heterobasidion annosum	-	0.1	-	-	0.1
Poria cinerascens	0.9	-	-	-	-
monokaryon	0.1	0.1	0.1	-	-
Fomitopsis cajanderi(monokaryon)	-	-	-	-	0.1
Phlebia albida	-	-	-	-	0.1
Unidentified basidiomycetes	0.9	0.2	1.1	0.7	0.6
Unidentified suspect fungi[b]	1.2	0.9	1.1	1.0	0.7
Number of cores with fungi[c]	140	81	68	84	54
Total cores taken	1952	1610	979	997	989
Percent cores with fungi	7.2	5.0	7.0	8.4	5.5

a. Not a basidiomycete.
b. Suspect fungi are those isolates which have basidiomycetous characteristics but lack clamp connections.
c. Does not equal the sum of the column as cores may have more than one fungus.

months exposure, probably due to competition from other basidiomycetes and imperfect fungi for readily available carbohydrates in the sapwood where Peniophora spp. were usually found (Chap. 3). The ratio of monokaryons to dikaryons generally was lower from pole sections exposed for 6 months, probably because dikaryons were formed by the monokaryons as they encountered compatible hyphae in the wood. The relatively high monokaryon isolation frequencies suggests high spore concentrations in the plots.

The use of exposed pole sections to estimate the inoculum levels of basidiomycetes has both positive and negative aspects. The fungi isolated are those whose propagules landed, germinated, and established colonies. Propagule viability, competition between fungi and fungal growth rate in the wood, weather conditions, and growth rate on malt medium all affect isolation frequencies. Thus, the basidiomycete isolation frequencies may not reflect the actual population of fungal propagules in the plots. However, the methods used during this study closely approximated actual air-seasoning conditions; therefore, the results are more meaningful than total spore counts would have been.

Because it was impossible to distinguish between expansion of existing decay-fungus colonies and newly established colonies in unsterilized pole sections, the

influence of sterilization on the number and species of decay fungi isolated was difficult to determine. One method of distinguishing between new and old colonies might involve a mating-type analysis of basidiomycetes isolated initially compared with those isolated after exposure. Monokaryons of each fungus would be crossed with monokaryons from all other cultures of the same species. Analysis of the incompatibility patterns would identify cultures with different genetic backgrounds. While this method would allow separation of genetically different isolates, different colonies arising from basidiospores from the same inoculum source could not be separated.

The variations in basidiomycete isolation frequency between plot locations suggests that sanitation measures aimed at reducing basidiospore inoculum levels could reduce pole colonization. These measures might include: using treated stickers and skids, removing all wood debris adjacent to air-seasoning poles, avoiding bulk log storage adjacent to the seasoning yard, and promptly removing cull poles. These measures are especially critical during the winter months, when the conditions are most favorable for wood colonization by decay fungi. Air-seasoning yards should be designed to reduce moisture levels at the soil surface as much as possible by graveling, providing

drainage, and avoiding areas with impeded air circulation.

Because environmental conditions during Pacific Northwest winters favor colonization of utility poles by decay fungi, protecting poles during this period could reduce pretreatment decay. Untreated poles exposed in the air-seasoning yards could be treated with standard wood preservatives or fungicides that prevent decay-fungus spore germination. Preliminary tests with ammonium bifluoride show promise for this use (35,65). Furthermore, pole inventories in air-seasoning yards could be decreased in the autumn to minimize the risk of decay during the winter.

CHAPTER 3

Frequency of Basidiomycetes in Douglas Fir Poles Air Seasoned in the Pacific Northwest

INTRODUCTION AND LITERATURE REVIEW

Decay of poles in service caused by basidiomycetes that colonize wood before preservative treatment has been suggested (10), and several studies support this hypothesis (16,104). These studies, and changes in treatment methods that may allow decay fungi in poles to survive preservative treatments already have been discussed (general introduction).

Because of relatively low cost, air seasoning is widely used in the Pacific Northwest to dry Douglas-fir poles prior to treatment. However, the time required to adequately air season poles may allow decay fungi to become established in the wood. Although little is known about colonization of Douglas-fir poles in air-seasoning yards, air seasoning southern yellow pine poles, even for a few weeks, may allow mold, stain and decay fungi to become established in the wood (65).

Colonization and decay studies of Douglas-fir logs in British Columbia (72,89) showed significant colonization by decay fungi after a 2 year exposure in the forest. A survey of log yards in the Pacific Northwest (57) found decay evident in Douglas-fir logs bulk-piled for 12 months.

Interest in pretreatment decay has been stimulated by a recent paper (94) that emphasized this problem in Douglas-fir poles. Strict specifications were recommended including sterilizing poles, banning of air-seasoned poles, and requiring Douglas-fir poles to be preservative treated within 3 months of cutting. It was also suggested that Douglas-fir poles were colonized by decay fungi within 3 to 6 months of cutting and that significant decay occurred within 6 to 12 months. However, none of these suggestions were supported by research on colonization of air-seasoning Douglas-fir poles.

The use of air seasoning as an energy-efficient method to dry poles prior to preservative treatment is increasing, and knowledge of pole colonization by decay fungi is urgently needed to evaluate the influence of this drying method on subsequent decay development.

The objectives of this study were: (i) to determine the frequency and identity of the most prevalent basidiomycetes in air-seasoning Douglas-fir poles, (ii) to determine the population changes of the basidiomycetes in poles as air-seasoning exposure increased, and (iii) to determine the distribution of the basidiomycetes within the poles.

MATERIALS AND METHODS

Air-seasoning poles in 24 yards in the Pacific Northwest were sampled during the summers of 1981 and 1982. Wood cores were removed at seven positions along the length of each pole: at each end, 2 and 4 feet in from each end, and in the middle of the pole. At each position two cores (6 inches long) were removed about 60 degrees apart for a total of 14 cores per pole. This pattern varied at times due to accessibility of poles in a pile.

Two increment borers and a power drill with a custom made adapter for the bits were used for sampling. The borers were used alternately, while a core was being removed from one, the other borer was used to obtain a core from a new location. In this manner cores could be taken rapidly by two people. The cores were processed and the basidiomycetes were isolated and identified as described in the general methods.

The unidentified fungi were separated into basidiomycetes and suspect fungi which have basidiomycetous characteristics, but lack clamp connections. The terms "basidiomycetes" and "decay fungi" in this chapter refer to all the basidiomycetes and suspect fungi isolated from the poles.

Sampled poles were placed arbitrarily in age classes depending on length of air seasoning. The age classes

have a degree of uncertainty because records of the origin and date of cutting of each pole were generally not kept by the pole owners. The age classes were: (i) fresh poles sampled within 4 weeks of cutting, mostly in the forest near the site of felling; (ii) unpeeled poles with bark intact sampled at the yard; and peeled poles in air seasoning for: (iii) 0 to 6 months; (iv) 7 to 12 months; (v) 13 to 18 months; (vi) 19 to 24 months and (vii) 25 months or more. Poles from northern California to northern Washington were sampled, in one or more age classes in each yard, depending on the current inventory and pole accessibility.

RESULTS AND DISCUSSION

The influence of borer temperature on the isolation frequency of basidiomycetes from wood

Using a power drill for core removal made possible the large numbers of poles sampled during this study, but borers were run at high speeds, and repeated use of the same borer heated the metal considerably. Because cores sometimes remained in the borers for several minutes, the heat could have affected the fungal population in the wood.

To determine if the heat generated during sampling introduced a bias, three pole sections exposed for 1 year at the Northwest Forest Genetics Station, Corvallis, OR were sampled using borers turned by a power drill or by

hand. Cores were taken in rows spaced 4 inches around the pole circumference and every 6 inches along the row. One set of cores was taken using a power drill and two borers as per usual. A second set was taken by hand, spaced 1 inch away from the power drill holes using eight borers which were cooled over dry ice between each use. The cores removed by both methods were processed, and the basidiomycetes were isolated from the cores, using the same methods used for cores from air-seasoning poles.

The isolation frequency of basidiomycetes was consistently higher in the cores taken by hand (Table 18); however, the differences were not significant (see appendix for statistics). This indicates that the heat generated by the power drill sampling did not significantly influence the results of sampling air-seasoning poles.

Changes in the populations of basidiomycetes during air seasoning of poles

As air-seasoning time increased, an increasing wood volume and number of poles colonized by basidiomycetes also increased (Fig. 3). The general trend in volume of wood colonized was obtained by summing the number of cores with basidiomycete isolates in each age class and expressing these sums as a percent of the total number of cores taken in each age class. The frequency of basidiomycetes expressed as percent cores colonized gives an

Table 18

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM CORES TAKEN BY HAND OR POWER DRILLING OF THREE DOUGLAS-FIR POLE SECTIONS

	Number of cores with basidiomycetes[a]	
Fungus species	Hand Cored	Power Cored
Poria placenta	71	55
Poria carbonica	50	43
Haematostereum sanguinolentum	29	23
Stereum hirsutum	4	1
Coriolus versicolor	1	1
Poria xantha	1	0
Peniophora spp.	0	1
Unidentified basidiomycetes	2	2
Unidentified suspect fungi[b]	11	14

a. The results are based on 215 cores taken by hand and 215 cores taken by power drilling.
b. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.

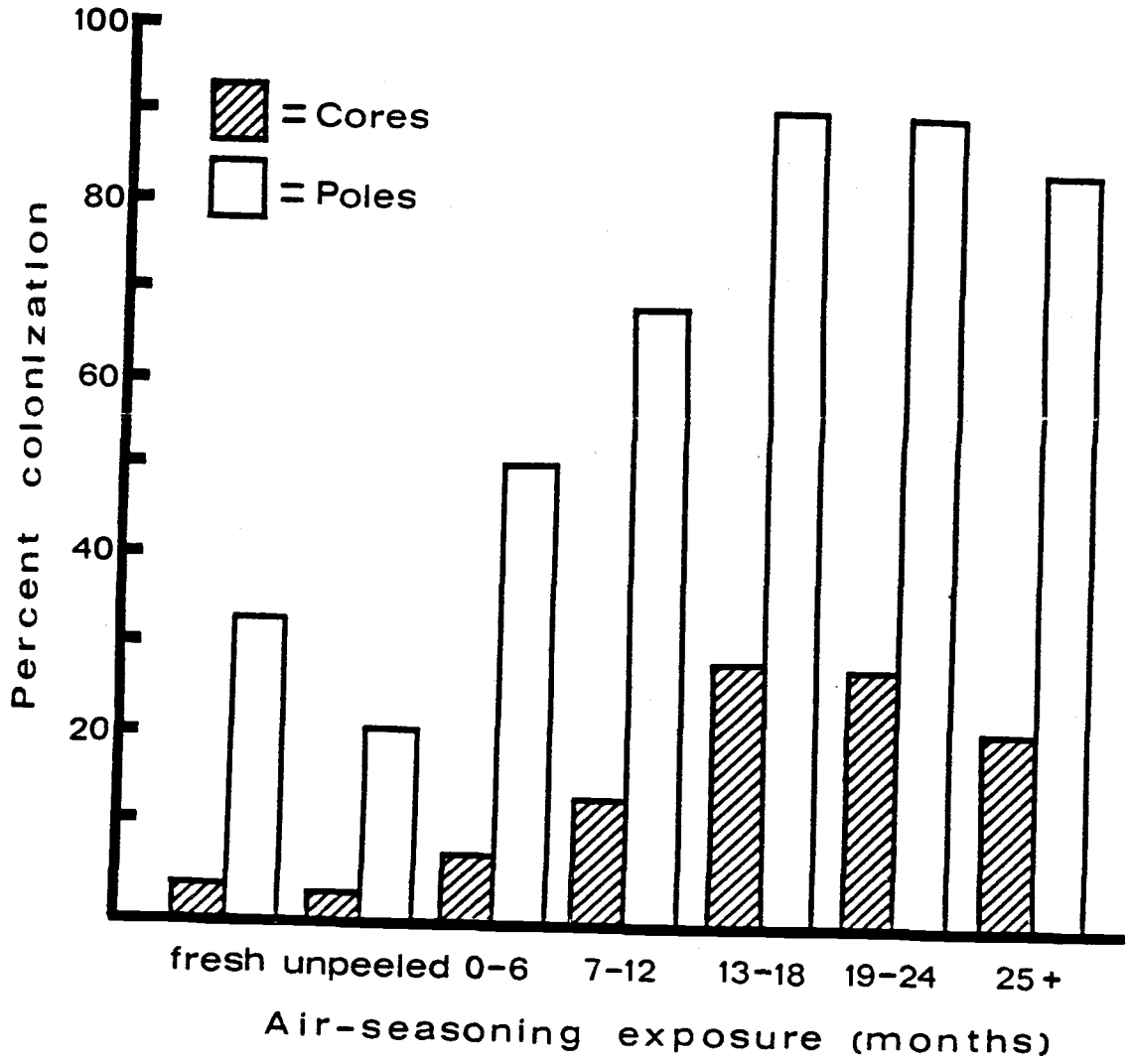


Fig. 3. Colonization of Douglas-fir poles air-seasoned for varying times in the Pacific Northwest expressed as percent poles and percent cores colonized by basidiomycetes. Total numbers of poles and cores sampled in each age class is given in tables 22 and 23, respectively.

estimate of the wood volume colonized, while percent poles colonized shows the distribution of decay fungi among poles.

Pole colonization by basidiomycetes occurred rapidly during the first 18 months of air seasoning (Fig. 3), then decreased slightly, probably due to depletion of readily available substrates in the sapwood. Pole and core colonization were similar, except in the fresh age class where the basidiomycetes were spread over a larger number of poles. Colonized poles in the 25+ month age class averaged about four colonized cores per pole, whereas freshly cut poles averaged less than one isolate per pole (Table 19).

Statistical comparison of basidiomycete isolation frequencies from poles in different age classes required an estimate of the variation between samples. In this study, each pole was considered a sample with the 14 cores from each pole as observations. Thus, comparisons of the average number of isolates per pole in each age class include variation between poles in each age class.

The average number of isolates per pole increased rapidly to a maximum of 4.7 at 18 months, then declined slightly (Table 19). Using this data, the seven arbitrary age classes could be placed into five distinct groups, based on the average number of isolates per pole (see appendix for statistics): (i) poles with their bark

Table 19

NUMBER OF BASIDIOMYCETES ISOLATED FROM DOUGLAS-FIR POLES AIR SEASONED FOR VARYING TIME PERIODS IN THE PACIFIC NORTHWEST

Number of cores with basidiomycetes per pole	Number of poles: Seasoning time (months) Fresh[a]	Unpeeled[b]	0-6	7-12	13-18	19-24	25+	Total
0	179	163	129	74	12	13	25	595
1	63	32	60	58	15	25	27	280
2	19	5	43	40	18	14	18	157
3	6	5	23	28	24	22	20	128
4	4	4	7	22	18	11	19	85
5	2	0	9	15	16	12	27	81
6	0	2	4	9	19	17	17	68
7	0	0	3	13	15	6	10	47
8	1	0	2	4	11	8	8	34
9	0	0	1	2	2	10	7	22
10	0	0	1	2	5	4	4	16
11	0	0	1	1	3	6	1	12
12	0	0	0	0	1	5	1	7
13	0	0	0	0	2	0	2	4
14	0	0	0	0	2	0	0	2
16	0	0	0	0	1	1	0	2
Total no. of cores with decay fungi	153	85	379	611	765	703	722	3418
Number of poles sampled	274	211	283	268	164	154	186	1540
Mean no. of cores with decay fungi per pole	0.6	0.4	1.3	2.3	4.7	4.6	3.9	
Standard deviation	1.0	1.0	1.9	2.4	3.2	3.5	3.0	
Similar groups[c]	A	A	B	C	D	D	E	

a. Sampled within 4 weeks of felling.
b. Sampled in pole yards with bark intact.
c. Age classes with same letter are not significantly different (see appendix for statistics).

intact, which included the fresh and unpeeled age classes, (ii) poles seasoned for 0 to 6 months, (iii) poles seasoned for 7 to 12 months, (iv) poles seasoned for 13 months to 24 months, and (v) poles seasoned for 25 months or longer. Thus, air seasoning Douglas-fir poles for more than 1 year, which is generally required to dry larger poles, ensures that a high percentage of these poles will contain basidiomycetes by the end of the seasoning.

The unpeeled poles were sparsely colonized, but after peeling the decay fungus population increased dramatically (Table 19). It appears that the exposure of unprotected sapwood, rich in soluble nutrients, resulted in a rapid colonization that declined as these nutrients became limiting at about 12 months. The ability to rapidly utilize these non-structural carbohydrates may determine if a decay fungus can successfully colonize the wood (42).

The influence of air-seasoning location on colonization of Douglas-fir poles by basidiomycetes

The frequency of basidiomycete isolation from air-seasoning poles varied considerably between yards, especially in the younger age classes (Table 20). For example, the percentage of "unpeeled" poles containing basidiomycetes in different yards ranged from 3 to 100%; suggesting that pole source, time in storage, and conditions between cutting and delivery to the yard may greatly influence colonization of unpeeled poles. Prompt

Table 20

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM DOUGLAS-FIR POLES AIR SEASONED FOR VARYING TIME PERIODS IN POLE YARDS IN THE PACIFIC NORTHWEST

Percent poles colonized by basidiomycetes

	Seasoning time (months)[a]						
Yard number	Fresh[b]	Unpeeled[c]	0-6	7-12	13-18	19-24	25+
1	10	-	-	-	-	-	-
2	51	-	-	-	-	-	-
3	24	-	-	-	-	-	-
4	26	-	-	-	-	-	-
5	50	-	-	-	-	-	-
6	55	-	-	-	-	-	-
7	-	25	74	74	-	-	65
8	-	-	28	88	-	87	-
9	-	6	35	25	-	-	90
10	-	100	85	95	100	-	-
11	-	-	-	55	67	-	68
12	-	-	-	-	75	-	-
13	-	50	55	-	91	100	100
14	-	-	-	-	-	100	100
15	-	3	53	0	97	85	100
16	-	-	-	-	96	-	-
17	-	-	-	-	-	95	-
18	-	-	-	-	100	-	100
19	-	7	29	53	-	76	-
20	-	-	-	-	-	89	-
21	-	25	25	65	-	-	100
22	-	-	-	-	-	100	100
23	-	16	86	84	100	-	-
24	-	26	66	90	-	95	85
Number of yards in age class	6	9	10	9	8	9	10
Average percent poles with decay fungi	36.1	28.6	53.6	69.8	90.6	91.8	90.8
Similar groups[d]	A	A	A	A	B	B	B

a. "-"= There were no poles sampled at this yard
b. Sampled within 4 weeks of felling.
c. Sampled in the yard with bark intact.
d. Age classes with the same letter not significantly different by (see appendix for statistics).

removal and transport of poles from the forest to the yard should decrease the probability of pole colonization before peeling.

Variation in pole colonization between yards decreased with increased air-seasoning time probably because of the heavy infestation of nearly all poles in the older age classes. This suggested that poles were being colonized by basidiomycetes in all the yards studied. These results indicate that inoculum levels of wood-decaying basidiomycetes are high throughout the Douglas-fir pole producing region in the Pacific Northwest. Because of the long rainy season and a relatively short summer, which limits pole drying, this region is not well-suited for air seasoning poles. The location of air-seasoning yards east of the Cascade mountains would probably speed drying and reduce the risk of pole colonization by basidiomycetes; however, increased transportation costs might make kiln or Boulton drying cost-effective.

Poles from both the Coast Range and the western slopes of the Cascade mountains were sampled during this study, but unfortunately, these poles were mixed in the seasoning yards and there was no way to differentiate between them. However, fresh-cut poles sampled in the forest could be identified. Three locations in the Coast Range were sampled and of 140 poles, 30.3% contained basidiomycetes.

Of 134 poles from three locations in the Cascade mountains, 41.7% were colonized by basidiomycetes. Pole colonization by basidiomycetes in these two distinct areas were not significantly different (Student's t-test, p=0.05), which suggests that the pole source has little effect on pole colonization by decay fungi.

Occurrence of different basidiomycete species in air-seasoning poles

The basidiomycete species isolated from air-seasoning poles changed dramatically as air-seasoning time increased (Table 21). Although P. carbonica and P. placenta account for most of the decay in Douglas-fir poles in service, these two species were not the basidiomycetes most frequently isolated from air-seasoning poles. While P. carbonica was infrequently isolated from poles early in air seasoning, it steadily increased after the 1st year to become the second most prevalent basidiomycete in poles air seasoned for 25 months or longer. P. placenta followed a similar trend, but at a lower level of wood colonization.

Most of the decay fungi isolated from air-seasoning Douglas-fir poles also have been isolated from other substrates. Poria carbonica and P. placenta are important causes of decay of Douglas-fir in poles, pilings, wooden boats and lumber (7,10,12,14). P. carbonica was the most frequently isolated decay fungus from Douglas-fir poles in

service in the Pacific Northwest (16) accounting for 56% of the decay fungi isolated, while Poria placenta was the second most prevalent isolate. A similar pattern of occurrence of these species was found in Douglas-fir poles in service in the Northeast (104).

Haematostereum sanguinolentum was frequently isolated from air-seasoning poles and its isolation frequency increased with air-seasoning time to a maximum at 13 to 18 months, after which it remained relatively constant. H. sanguinolentum is a white rot fungus that causes a heartrot in living conifers and continues to decay the wood after the tree is cut (37,40,41). This fungus causes 72% of the decay initiated in thinning wounds in Douglas-fir (41), and is commonly found on slash and recently downed trees. H. sanguinolentum also has been reported on stored pulp, lumber, and was associated with death of planted conifer seedlings (8,10,14,40). This fungus has not been reported from poles in service, probably because it is mostly in the sapwood where it is killed during preservative treatment.

The distribution of Coriolus versicolor between the age classes was relatively even, except for the 13 to 18 month class in which its high frequency was due to a 30 pole sample from one yard in which 15 poles were infested with this fungus. C. versicolor causes a soft white spongy rot of dead sapwood, and a white rot of heartwood

Table 21

FREQUENCY OF BASIDIOMYCETES COMMONLY ISOLATED FROM DOUGLAS-FIR POLES AIR SEASONED FOR VARYING TIME PERIODS IN THE PACIFIC NORTHWEST

	Percent poles colonized by basidiomycetes							
			Seasoning time (months)[a]					
Fungus species[b]	Fresh[c]	Unpeeled[d]	0-6	7-12	13-18	19-24	25+	Total
Haematostereum sanguinolentum	1	2	19	26	51	45	48	24
Peniophora spp.	0	0	5	25	49	37	21	17
Sistotrema brinkmanii[e]	1	2	12	19	19	21	20	12
Poria carbonica	T	0	1	9	19	27	39	11
monokaryon	0	0	T	1	2	3	3	1
Epicoccum nigrum[f]	T	4	7	12	24	24	10	10
Poria placenta	1	1	2	8	13	18	16	7
monokaryon	1	0	2	3	7	8	9	4
Coriolus versicolor	6	2	5	3	15	5	8	6
monokaryon	4	1	3	2	2	1	3	2
Stereum hirsutum	1	1	2	5	11	8	5	4
Unidentified basidiomycetes	4	4	7	22	35	32	35	18
Unidentified suspect fungi[g]	20	8	11	6	20	12	12	12
Percent poles with fungi	34.7	22.8	54.4	72.4	92.7	91.6	86.6	61.4
Total number of poles sampled	274	211	283	268	164	154	186	1540
Total number of cores taken	3834	2528	3939	3784	2350	2159	2628	21222

a. "T" = less than 0.5%.
b. The monokaryons are included with the dikaryons and are also shown separately to give a breakdown between dikaryon and monokaryon.
c. Sampled within 4 weeks of felling.
d. Sampled in the yard with the bark intact.
e. This is a species complex.
f. A non-basidiomycete fungus that may influence wood strength.
g. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.

(8). This widely distributed fungus attacks wood in storage and in service, is common on hardwood slash (8,12,14,101), and has been reported in treated southern pine and Douglas-fir poles (104,105).

The distributions of Peniophora spp., Sistotrema brinkmanii and Epicoccum nigrum were similar between age classes of air-seasoned Douglas-fir poles. They increased to a maximum frequency after 1 year, then decreased suggesting that these species were utilizing the easily accessible carbohydrates in the sapwood. Both Peniophora spp. and S. brinkmanii produce sporophores on the sapwood of untreated poles in air-seasoning yards (46).

Peniophora spp. represents a group of white rot fungi that are difficult to separate into individual species. They are common in conifer sapwood, and have been reported from lumber, railroad ties and poles (10,12,14,16). These fungi, although capable of decaying wood, are generally limited to the sapwood (10,14,16).

Sistotrema brinkmanii is a brown rot fungus that is common in soil and decaying slash (56,92), and has been isolated from southern pine poles (18,105). This fungus is capable of causing decay in ponderosa pine stakes after 12 weeks (56), and may be causing strength loss in Douglas fir poles.

Epicoccum nigrum is a non-basidiomycete soft-rot fungus found in treated southern pine poles (105), where

it may cause some strength loss.

Twenty-two additional basidiomycetes were isolated from air-seasoning Douglas-fir poles (Tables 22, 23), and the greatest variety of species were isolated during the first 6 months of air seasoning. After this initial period, the decreasing species numbers were probably caused by increased competition among basidiomycetes and imperfect fungi for available nutrients. Many of these additional decay fungi have been reported from poles, lumber and other wood products, where they may cause serious problems (8,10,12,14,16,104). However, these species were isolated at relatively low frequencies from the air-seasoning poles and probably do not pose serious problems. But as treatment practices change, the basidiomycete species causing decay in poles in service may also change. Some decay fungi isolated in low frequencies could cause decay problems in poles treated with some preservative treatments, especially if they survived the initial treatment.

Colonization of air-seasoning Douglas-fir poles by monokaryotic basidiomycetes

The techniques used in this study did not allow differentiation between expansion of existing fungal colonies in wood and establishment of new colonies. Consequently, the increased isolation frequency of basidiomycetes from poles as air-seasoning time increased

Table 22

FREQUENCY OF BASIDIOMYCETES ISOLATED FROM DOUGLAS-FIR POLES AIR SEASONED FOR VARYING TIME PERIODS IN THE PACIFIC NORTHWEST

Percent poles colonized by basidiomycetes

	Seasoning time (months)							
Fungus species	Fresh[a]	Unpeeled[b]	0-6	7-12	13-18	19-24	25+	Total
Haematostereum sanguinolentum	1.1	1.9	19.1	25.4	51.2	44.8	48.4	24.2
Peniophora spp.	0	0	4.6	24.6	49.4	36.6	21.0	16.8
Sistotrema brinkmanii[c]	1.1	1.9	12.0	18.7	18.9	21.4	20.4	12.5
Poria carbonica	0.4	0	1.1	7.8	16.5	23.4	36.6	10.1
Epicoccum nigrum[d]	0.4	3.8	6.7	11.6	23.8	24.0	9.7	9.9
Phanerochaete sordida	5.1	0.5	2.1	2.2	9.1	11.0	5.2	4.2
Stereum hirsutum	1.1	0.9	1.8	5.2	11.0	8.4	5.4	4.2
Poria placenta monokaryon	0.8	0	2.1	3.4	6.7	8.4	9.1	3.8
Poria placenta	0	0.9	0.4	4.9	6.7	9.7	6.5	3.5
Coriolus versicolor	1.8	1.4	2.1	1.1	12.8	3.9	4.8	3.4
Coriolus versicolor monokaryon	2.9	0.9	3.2	1.5	1.8	1.3	2.7	2.1
Gloeophyllum saeparium	0.4	0.5	0.7	0	3.0	6.5	7.5	2.1
Fomitopsis cajanderi	0.4	2.4	1.4	2.2	1.8	1.3	1.6	1.6
Schizophyllum commune	0.4	0.5	1.1	0.4	6.1	1.9	1.1	1.4
Poria carbonica monokaryon	0	0	0.4	0.7	2.4	3.2	2.7	1.1
Cystostereum pini-canadense	0	0	2.8	0	0	0	0	0.5
Phlebia "A" monokaryon	0.8	0.5	1.1	0.4	0.6	0	0	0.5
Schizophyllum commune monok.	0.8	0	1.1	0	1.8	0	0	0.5
Phlebia radiata monokaryon	0.4	0	0.4	0.4	1.2	0.6	0	0.4
Poria cinerascens monokaryon	0	0	1.4	0	1.2	0	0	0.4
Fomitopsis pinicola monokaryon	1.2	0.5	0.4	0	0	0	0.5	0.4
Heterobasidion annosum	0	0	0.4	0.4	0	0.6	0	0.2
Fomitopsis pinicola	0.4	0	0.4	0	0	0	0	0.1
Phlebia gigantea	0	0.5	0.4	0	0	0	0	0.1
Poria xantha	0	0	0	0	0	0	0.5	0.1
Poria cinerascens	0	0	0	0	0.6	0	0	0.1
Phlebia albida monokaryon	0	0	0	0	0	0.6	0	0.1
Crustoderma dryinum	0	0	0	0	0	0	0.5	0.1
Poria xantha monokaryon	0	0	0	0	0	0.6	0	0.1
Fomitopsis cajanderi monokaryon	0	0	0.4	0	0	0	0	0.1
Unidentified basidiomycetes	4.4	3.8	7.1	22.0	34.8	32.5	34.9	17.6
Unidentified suspect fungi[e]	20.4	7.6	11.0	5.6	20.1	11.7	12.4	12.5
Total number of poles with basidiomycetes[f]	95	48	154	194	152	141	161	945
Total number of poles sampled	274	211	283	268	164	154	186	1540

a. Sampled within four weeks of felling.
b. Sampled in the yard with the bark intact.
c. This is a species complex.
d. A non basidiomycete fungus that may influence wood strength.
e. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
f. Does not equal the sum of the column as one core may have more than one fungus.

Table 23

FREQUENCY OF BASIDIOMYCETES ISOLATED IN CORES FROM DOUGLAS-FIR POLES AIR SEASONED FOR VARYING TIME PERIODS IN THE PACIFIC NORTHWEST

Number of cores colonized by basidiomycetes

	Seasoning time (months)							
Fungus species	Fresh[a]	Unpeeled[b]	0-6	7-12	13-18	19-24	25+	Total
Haematostereum sanguinolentum	3	7	116	153	193	191	205	868
Peniophora spp.	0	0	14	93	176	110	58	451
Sistotrema brinkmanii[c]	3	4	70	108	41	51	69	346
Poria carbonica	1	0	5	31	36	64	124	260
Epicoccum nigrum[d]	1	9	22	44	51	63	26	216
Fomitopsis cajanderi	3	18	10	19	8	11	13	82
Coriolus versicolor	5	3	6	6	39	7	12	78
Phanerochaete sordida	15	1	6	6	16	21	7	72
Stereum hirsutum	3	2	5	14	21	15	10	70
Poria placenta monokaryon	2	0	7	9	13	21	18	70
Poria placenta	0	2	1	17	14	19	15	68
Gloeophyllum saeparium	1	1	2	0	5	17	17	43
Coriolus versicolor monokaryon	11	2	9	4	3	2	5	36
Schizophyllum commune	1	1	9	2	17	3	2	35
Poria carbonica monokaryon	0	0	1	2	4	5	5	17
Cystostereum pini-canadense	0	0	13	0	0	0	0	13
Phlebia "A" monokaryon	2	1	3	1	1	0	0	8
Schizophyllum commune monok.	2	0	3	0	3	0	0	8
Phlebia radiata monokaryon	1	0	1	1	2	1	0	6
Poria cinerascens monokaryon	0	0	4	0	2	0	0	6
Fomitopsis pinicola monokaryon	3	1	1	0	0	0	1	6
Heterobasidion annosum	0	0	1	1	0	1	0	3
Fomitopsis pinicola	1	0	2	0	0	0	0	3
Phlebia gigantea	0	1	1	0	0	0	0	2
Poria xantha	0	0	0	0	0	0	1	1
Poria cinerascens	0	0	0	0	1	0	0	1
Phlebia albida monokaryon	0	0	0	0	0	1	0	1
Crustoderma dryinum	0	0	0	0	0	0	1	1
Poria xantha monokaryon	0	0	0	0	0	1	0	1
Fomitopsis cajanderi monokaryon	0	0	3	0	0	0	0	3
Unidentified basidiomycetes	14	13	25	82	81	82	111	408
Unidentified suspect fungi[e]	85	19	40	19	39	23	24	249
Total number of cores with basidiomycetes[f]	150	84	372	591	722	643	667	3229
Total number of cores taken	3834	2528	3939	3784	2350	2159	2628	21222

a. Sampled within four weeks of felling.
b. Sampled in the yard with the bark intact.
c. This is a species complex.
d. A non basidiomycete fungus that may influence wood strength.
e. Suspect fungi are those isolates which have basidiomycetous characteristics, but lack clamp connections.
f. Does not equal the sum of the column as one core may have more than one fungus.

was due to both expansion of existing colonies and new colonies arising from fungal propagules that landed on the wood.

Colonies resulting from basidiospore germination on wood are generally monokaryotic, and their prevalence in air-seasoning poles suggests that decay fungi are initiating colonization in the air-seasoning yards (Table 21). Poria placenta and Coriolus versicolor monokaryons account for about one half of the total isolates of these fungi, suggesting high local spore populations. The low frequency of P. carbonica monokaryons was probably due to fewer basidiospores and a slow growth rate in wood.

In studies of Douglas-fir log deterioration (57,89), P. carbonica was isolated more frequently from logs exposed for longer times, further suggesting that P. carbonica does not successfully compete with other decay fungi in initial stages of wood colonization.

Basidiomycete distribution in air-seasoning Douglas-fir poles

More basidiomycetes were isolated from the outer shell of poles in all age classes than from the inner portions except at the pole ends (Fig. 4). The outer 2 inches and the inner 4 inches of the cores corresponded roughly to the sapwood and the heartwood, respectively. The frequency of basidiomycetes in the outer portions of the cores decreased significantly at the butt, probably due to

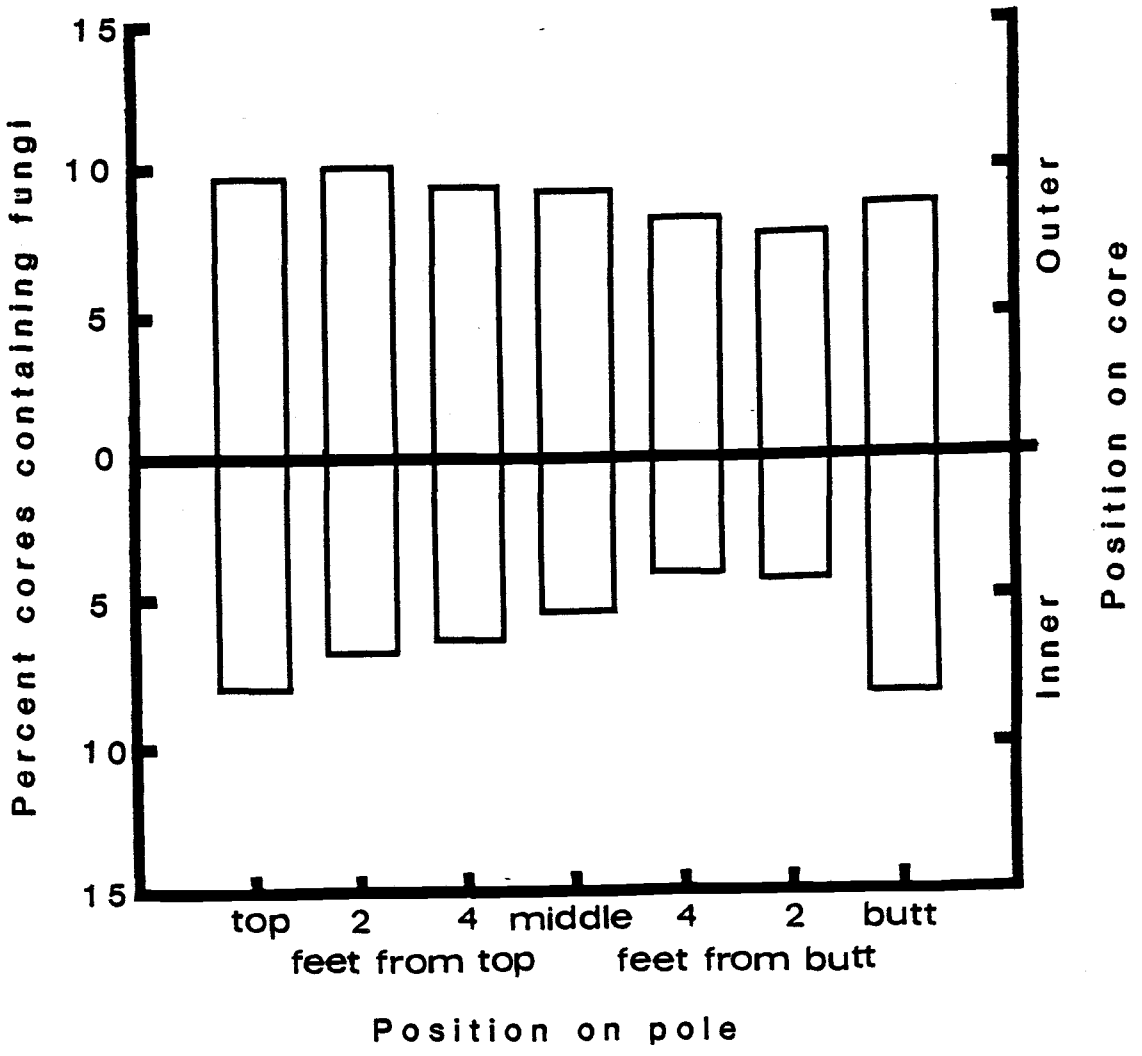


Fig. 4. Distribution of basidiomycetes along the length of Douglas-fir poles air-seasoned for varying time periods in the Pacific Northwest. The inner portion on the core is the inner 4 inches of each core, and is mostly heartwood. The outer portion of the core is the outer 2 inches of each core, and is mostly sapwood.

the removal of sapwood from the butt during peeling to reduce pole taper (see appendix for statistics).

Basidiomycetes isolated from the inner portion of the cores were concentrated at the pole ends. This was probably due to exposure of the heartwood which allowed easy access and also enhanced diffusion of oxygen and water.

The distribution of basidiomycetes on and along the pole changed with increasing air-seasoning time (Fig. 5). Peeling the poles exposed the sapwood, a nutrient rich substrate, and allowed the wood surface to dry. Drying caused checks to form, which are likely sites of colonization. As a result, there was rapid colonization by fast-growing basidiomycetes such as Schizophyllum commune and Peniophora spp. These fungi probably have little effect on pole strength (73), but they may modify the wood which allowed colonization by other decay fungi (6,67,68,70).

Basidiomycete frequency in the inner portion of poles in the 7-12 month age class was about the same as in the outer 2 inches of the poles, except for the butt end (Fig. 5). As seasoning time increased, basidiomycete frequency in the outer 2 inches of the poles, which was relatively uniform along the pole length, increased significantly

Fig. 5. Distribution of basidiomycetes in four age groups along the length of Douglas-fir poles air-seasoned for varying time periods in the Pacific Northwest. The inner core position was the inner 4 inches of each core, which was mostly heartwood. The outer core position was the outer 2 inches of each core, which was mostly sapwood. The four groups plotted are the age classes grouped by analysis of number of isolates per pole (Table 17).

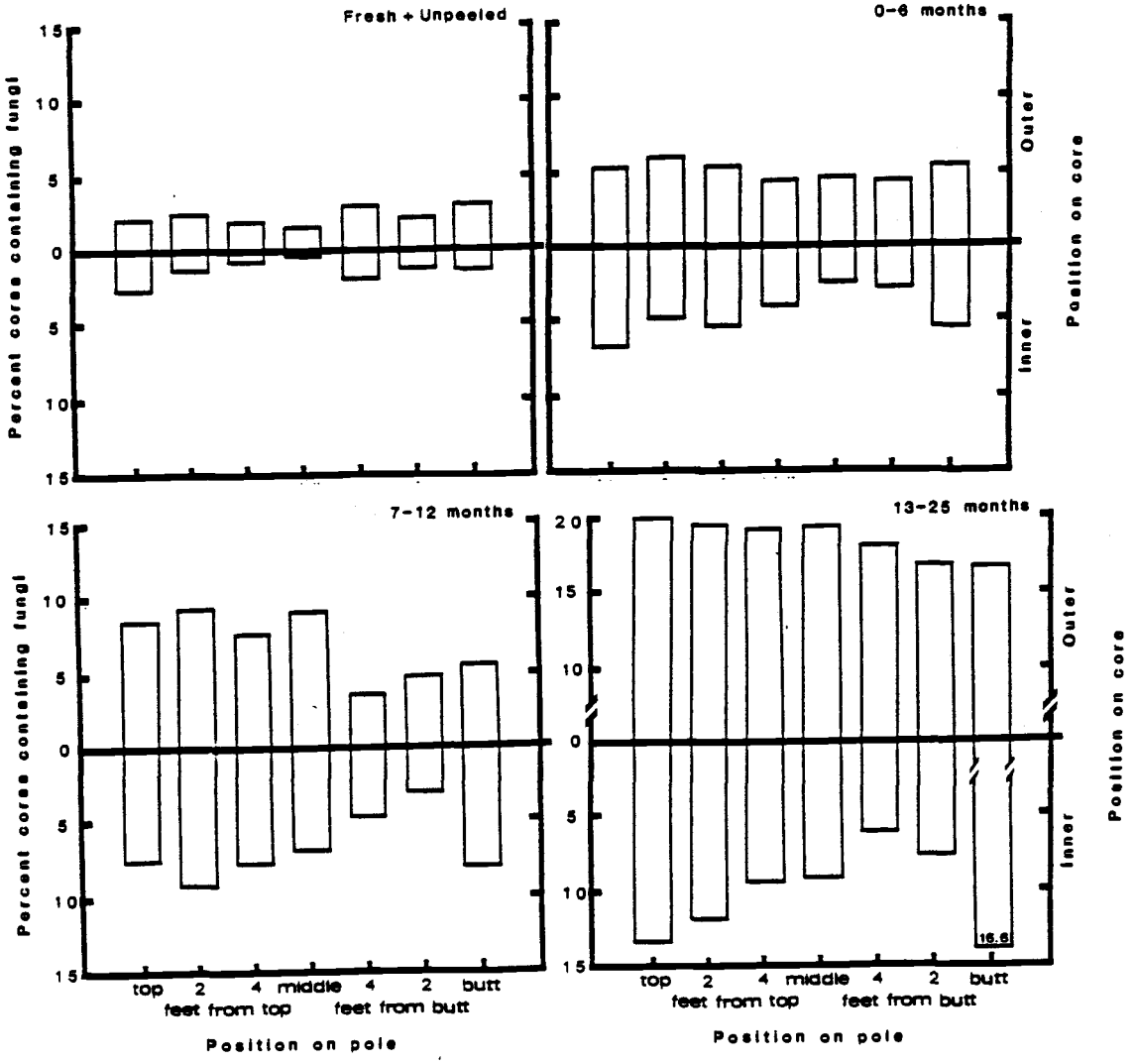

Fresh + Unpeeled
0-6 months
7-12 months
13-25 months
Percent cores containing fungi
Position on core
Outer
Inner
15
10
5
0
5
10
15
20
16.6
top
2
4
middle
4
2
butt
feet from top
feet from butt
Position on pole

Fig. 5.

more than that from the inner portion, except at the butt end.

The distribution of basidiomycete species along the pole length show distinct patterns (Fig. 6). Haematostereum sanguinolentum, Peniophora spp. and Sistotrema brinkmanii were the most prevalent basidiomycetes isolated in the 0-6 month age class (Table 21), and they were mostly in the sapwood (Fig. 6).

Poria carbonica and P. placenta were found most frequently in the inner portion of poles where P. carbonica was particularly concentrated in the butt ends, accounting for about 25% of the isolates from this end. These two fungi, which were isolated most frequently from the heartwood, probably have a competitive advantage in this zone which may partially explain their prevalence in Douglas-fir poles in service. The concentration of these two fungi in the inner portion of poles increases the chance of them surviving preservative treatment, and later causing decay of poles in service.

General discussion

The results of this study indicate that air seasoning Douglas-fir poles for more than 1 year assures that most of the poles will be colonized by basidiomycetes. As the seasoning time increased, the basidiomycete populations shifted from fast growing sapwood colonizers, to slower-growing heartwood colonizers capable of causing

Fig. 6. Distribution of four basidiomycetes along the length of Douglas-fir poles air-seasoned for varying time periods in the Pacific Northwest. The inner core position was the inner 4 inches of each core, which was mostly heartwood. The outer core position was the outer 2 inches of each core, which was mostly sapwood.

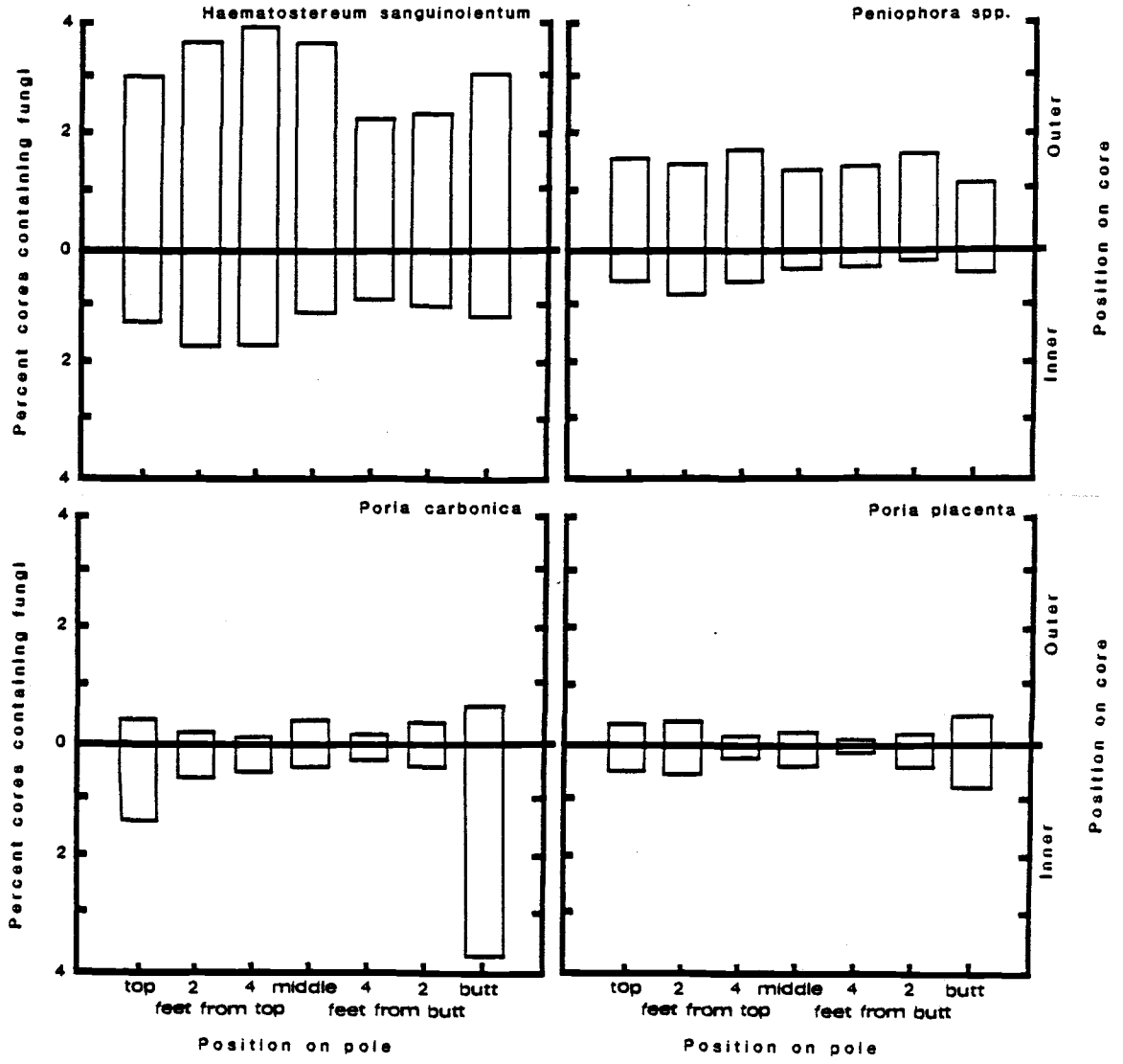

Haematostereum sanguinolentum
Peniophora spp.
Poria carbonica
Poria placenta
Percent cores containing fungi
4
2
0
2
4
Outer
Inner
Position on core
top 2 4 middle 4 2 butt
feet from top
feet from butt
Position on pole

Fig. 6.

significant decay in Douglas-fir. This pattern of fungal succession has been observed in other wood species (6,67,68,69,70).

Most of the basidiomycetes in the pole ends and the outer pole shell probably are killed during preservative treatment because chemical penetration is greatest at the ends and in the sapwood. Incising and drilling poles increases the depth of preservative penetration (27), producing a thicker treated shell.

Basidiomycetes isolated mostly from the heartwood, e.g Poria carbonica and P. placenta, were uncommon in poles seasoned less than 1 year. Thus, limiting air seasoning to less than 1 year may avoid some potential decay problems. Requiring longer heating times and higher temperatures during preservative treatment also would improve decay control. Another promising approach is the use of fumigants (31,105), which could be placed in the ground line area of poles during framing. These chemicals would eliminate basidiomycetes from poles in the area most prone to decay.

The high frequency of white-rotting fungi in air-seasoning poles is unusual in that these fungi generally do not cause significant problems in Douglas-fir poles. However, as treatment practices change to meet the demand for environmentally safe preservatives, the fungi causing

decay problems may also change. Additional research is needed to determine if basidiomycetes in Douglas-fir poles survive various types of preservative treatments.

Initial colonization of poles probably occurs on the exposed pole ends. Decay fungi colonize wood at a faster rate along the grain (99), which is why most of the wood samples were taken near the pole ends during this study. Each pole was sampled by removing 14 cores, representing less than 0.01% of the wood volume of a 40 foot pole, 12 inches in diameter. Taking more cores per pole might have changed the percentage of colonized poles and the estimates of the wood volume colonized by basidiomycetes.

The results may not represent the actual frequency of basidiomycetes in the entire pole because of the limited number of cores taken per pole. Estimates of the basidiomycetes in the heartwood may be high because sampling of the air-seasoning poles was concentrated at the ends. Decay fungi could have colonized the exposed heartwood at the end, and grown inward, while the heartwood in the middle of the poles remained relatively free of basidiomycetes. The ends of air-seasoned poles are trimmed prior to treatment which may remove much of the wood sampled during this study. Further sampling of freshly treated poles, especially near the ends, is needed to determine if these fungi survive.

A decay-development study was designed to determine

the distribution of decay fungi in pole section exposed for 1, 2 or 3 years in air-seasoning yards. After exposure each pole section was extensively sampled to determine the spatial distribution of decay fungi in the pole sections. Results of this study are pending identification of the decay fungi isolates and analysis.

GENERAL DISCUSSION

Comparing basidiomycete isolation frequencies from air-seasoning Douglas-fir poles with those from sterilized pole sections showed some interesting differences. Poria placenta was the most frequently isolated decay fungus from sterile pole sections exposed for 3 month intervals (Table 15), whereas this fungus was the eleventh most frequent in air-seasoning poles (Table 22). P. placenta inoculum levels are probably high in the air-seasoning yards, thus it could rapidly colonize the sterile pole sections with little competition from other decay fungi. However, other fungi present in Douglas-fir poles arriving at the pole yards probably competed for nutrients and limited colonization and growth of P. placenta in air-seasoning poles.

Conversely, Haematostereum sanguinolentum was the most frequently isolated decay fungus from air-seasoning poles (Table 22), but was the seventh most frequent from the sterilized pole sections exposed for 3 month intervals (Table 15). H. sanguinolentum was isolated in higher frequencies from pole sections exposed for 6 months (Table 16), suggesting that this fungus requires longer incubation periods. H. sanguinolentum has been isolated from living trees (41,42) and was isolated from fresh and unpeeled poles. This, coupled with the low isolation frequency from pole sections suggests that its high

isolation frequency from air-seasoning poles was partly due to expansion of colonies present in the wood at the time of cutting.

Sistotrema brinkmanii and Peniophora spp. were prevalent in both the sterile pole sections and air-seasoning poles and were more frequent in sterile pole sections exposed for 6 months than those exposed for 3 months. This suggests that these fungi are colonizing Douglas-fir poles in the air-seasoning yards.

The distribution of decay fungi in the sterilized pole sections was similar to that observed in air-seasoning poles (Table 17, Fig. 6). Poria placenta was isolated mostly from heartwood near the ends, while Peniophora spp. and H. sanguinolentum were isolated mostly from sapwood along the pole length.

Culturing wood cores taken with increment borers to isolate the decay fungi from poles has several drawbacks. Although the use of increment borers allowed large numbers of poles to be sampled, the borers were not sterilized between cores and decay fungi from one core may have been transferred to the next core taken. However, flaming of the cores prior to placing them in culture media should have killed any external contaminants.

The probability of detecting decay fungi present in low frequencies increases as larger volumes of wood are

cultured. But, because the entire core was placed in the medium, the possibility of one fungus inhibiting the growth of another fungus from the wood was increased. To limit this inhibition, the cores could have been cut into small chips before plating (104), however the time required would have severely limited the number of poles sampled during these studies.

The coring patterns used in sampling air-seasoning Douglas-fir poles were markedly different from those used in the infection study. The sterilized pole sections were more intensively sampled than the air-seasoning poles, which increased the probability of detecting small decay fungi colonies. Samples taken from air-seasoning poles were concentrated at the pole ends to increase the possibility of detecting decay fungi. However, isolation results from the 4-foot pole sections exposed horizontally (Table 17), indicate the upper surfaces of the poles are equally susceptible to basidiomycete colonization.

Although most poles air seasoned for more than 1 year are colonized by decay fungi, many of these fungi are in the sapwood and are killed during preservative treatment. Additional research is needed to determine the effects on pole strength caused by the most prevalent fungi isolated from air-seasoning Douglas-fir poles, and the effects of various pole treatments on the survival of these fungi in the finished poles.

BIBLIOGRAPHY

1. Amburgey, T.L. 1970. Relationship of capacity to cause decay to other physiological traits in isolates of Lenzites trabea. Phytopath. 60: 955-960.

2. ANSI. 1979. Specifications and dimensions for wood poles, 0.511979. American National Standards Institute, Inc, New York. 20p.

3. Badcock, E.C. 1941. New methods for the cultivation of wood-rotting fungi. Trans. Brit. Myc. Soc. 25: 200-205.

4. Badcock, E.C. 1943. Methods for obtaining fructifications of wood rotting fungi in culture. Trans. Brit. Myc. Soc. 26: 127-132.

5. Behr, E.A. 1973. Decay test methods. Chap. 6 in Nicholas, D.D. ed. Wood deterioration and its prevention by preservative treatments. Syracuse Univ. Press. Syracuse, NY.

6. Blanchette, R.A. and C.G. Shaw. 1978. Associations among bacteria, yeasts and basidiomycetes during wood decay. Phytopath. 68: 631-637.

7. Boyce, J.S. 1923. A study of decay in Douglas-fir in the Pacific Northwest. USDA Bull. 1163: 1-19.

8. Boyce, J.S. 1961. Forest Pathology McGraw-Hill Book Company, Inc. N.Y. 572 p.

9. Butcher, J.A., Hedley, M.E. and J. Drysdale. 1977. Comparison of a quaternary ammonium compound and copper-chrome-arsenate as wood preservatives. For. Prod. J. 27(7): 22-25.

10. Cartwright, K.T. and W.P.K. Findlay. 1958. Decay of Timber and Its' Prevention. Her Majesty's Stationary Office London. 332 p.

11. Chapman, A.D. 1933. Effect of steam sterilization on the susceptibility of wood to blue-staining and wood destroying fungi. Jour. Agr. Res. 47: 369-374.

12. Cowling, E.B. 1957. A partial list of fungi associated with decay of wood products in the U.S. Pl. Dis. Rep. 41: 894-896.

13. DaCosta, E.W.B. and R.M. Kerruish. 1965. The comparative wood-destroying ability and preservative tolerance of monokaryotic and dikaryotic mycelia of Lenzites trabea and Poria vaillantii. Ann. Bot. 29: 241-252.

14. Duncan, C.G. and F.F. Lombard. 1965. Fungi associated with decay in wood products in the U.S. USDA For. Ser. Res. Paper. WO-4. 31 p.

15. Elliot, C.G., An Abou-Heilah, Leake, D.L. and S.A. Hutchinson. 1979. Analysis of wood decaying ability of monokaryons and dikaryons of Serpula lachrymans. Trans. Brit. Myc. Soc. 73(1): 127-133.

16. Eslyn, W.E. 1970. Utility pole decay. Part II. Basidiomycetes associated with decay in poles. Wood Sci. and Tech. 4: 97-103.

17. Ettner, B. 1928. New media for developing sporophores of wood rotting fungi. Mycologia. 21: 197-203.

18. Frankland, J.C., Hedger, J.N. and M.J. Swift. editors. 1982. Decomposer basidiomycetes: their biology and ecology. Cambridge Univ. Press. Cambridge, Eng.

19. Gibson, I.A.S. and J. Trapnell. 1957. Sporophore production by Polyporous arcularius in culture. Trans. Brit. Myc. Soc. 40(2): 213-220.

20. Gilbertson, R.L. 1981. North American wood-rotting fungi that cause brown rots. Mycotaxon. 12: 372-416.

21. Gillette, D.A. 1974. On the production of soil wind erosion aerosols having the potential for long range transport. Jour. Rech. Atmosph. 8: 735-744.

22. Gillette, D.A., Blifford, I.A. and D.W. Fryrear. 1974. Wind erosion aerosols of soils. Jour. Geophy. Res. 79(27): 4068-4075.

23. Good, H.M. and W. Spanis. 1958. Some factors affecting the germination of spores of Fomes igniarius var. populinus (Neuman) Campbell, and the significance of these factors. Can. Jour. Bot. 36: 421-437.

24. Gottlieb, D. 1950. The physiology of spore germination in fungi. The Bot. Rev. 16(5): 229-257.

25. Graham, R.D. 1984. personal comm.

26. Graham, R.D. 1983. Improving the performance of wood poles. Proc. Amer. Wd. Pres. Assn. 79:222-228.

27. Graham. R.D. 1980. Converting 1979 wood preservation problems into opportunities for 1999. For. Prod. Jour. 30(2): 17-20.

28. Graham, R.D. 1980. Boulton drying; a review of its effects on wood. Appendix B. Report of committee T-4. Proc. Amer. Wood Pres. Assn. 76: 85-88.

29. Graham, R.D. and M.E. Corden. 1980. Controlling biological deterioration of wood with volatile chemicals. Final report. Electric Power Research Institute. EL-1480 RP212-1. Palto Alto, CA.

30. Graham, R.D. and R.J. Womack. 1972. Kiln and boulton drying Douglas fir pole sections at 220 to 290°F. For. Prod. Jour. 22(10): 50-55.

31. Graham, R.D. and G.G. Helsing. 1979. Wood pole maintenance manual: inspection and supplemental treatment of Douglas-fir and western redcedar poles. Forest Research Laboratory. Oregon St. Univ. Corvallis. Res. Bull. 24. 64p.

32. Gram, G. 1979. Fruiting in higher fungi II. Wood destroying basidiomycetes. Zeitschrift fur Mykologie. 45(2): 195-208.

33. Hawley, L.F. and J. Wiertelak. 1931. Effect of mild heat treatments on the chemical composition of wood. Jour. Ind. & Eng. Chem. 23(2): 184-186.

34. Helsing, G. and R.D. Graham. 1976. Saw kerfs reduce checking and prevent internal decay in pressure treated Douglas-fir poles. Holzforschung. 30: 184-186.

35. Helsing, G. and R.D. Graham. 1980. Protecting cutoff tops of Douglas-fir piles from decay. For. Prod. J. 30(2): 23-25.

36. Henry, W.T. 1973. Treating processes and equipment. in Wood deterioration and its prevention by preservative treatment. D. Nicholas ed. Vol. II. Preservatives and preservative systems. Syracuse Univ. Press.

37. Hepting, G.H. 1971. Diseases of forest and shade trees of the United States. USDA Handbook No. 386.

38. Hintikka, V. 1970. Stimulation of spore germination of wood-decomposing hymenomycetes by CO_2. Karstenia. XI: 23-27.

39. Hirst, J.M., Stedman, O.J. and W.H. Hogg. 1967. a. Long distance spore transport: methods of measurement, vertical spore profiles and the detection of immigrant spores. J. Gen. Microbiol. 48: 329-355.

40. Hubert, E.E. 1935. A disease of conifers caused by Stereum sanguinolentum. Jour. of For. 33: 485-489.

41. Hubert, E.E. and K.W. Krueger. 1962. Decay associated with thinning wounds in young-growth western hemlock and Douglas fir. Jour. of For. 60: 336-340.

42. Hulme, M.A. and J.K. Shields. 1970. Biological control of decay fungi in wood by competition for non-structural carbohydrates. Nature. 227:300-301.

43. Hunt, G.H. and G.A. Garratt. 1953. Wood preservation. McGraw Hill Book Co. Inc. N.Y. 417p.

44. Kallio,T. 1971. Aerial distribution of some wood-inhabiting fungi in Finland. Acta Forestalia Fennica 115. 17p. Helsinki.

45. Kozlik, C.J. 1982. Kiln-drying Douglas fir pole sections: effects on strength and checking. For. Prod. Jour. 32 (6): 24-30.

46. Kropp, B. 1984. personal comm.

47. Larsen, H.J. and R.P. Covey. 1979. A rapid slide mount technique for agar grown fungal cultures. Phytopath. 69(6): 682-683.

48. Li, C.Y. 1979. Light and temperature induced sporocarp production of Phellinus weirii. Can. Jour. For. Res. 9(4): 535-538.

49. Long, W.H. and R.M. Harsch. 1918. Pure cultures of wood-rotting fungi on artificial media. J. Agr. Res. XII(2): 33-82.

50. Lowe, J.L. 1966. Polyporaceae of North America. The genus Poria. St. Univ. Coll. For. Syracuse Technical Publication No. 90. 183 p.

51. Lu, B.C. 1974. The role of light on basidiocarp initiation, mitosis and hymenium differentiation in Coprinus lagopus. Can. J. Bot. 52(2): 299-305.

52. Manion, P.D. and D.W. French. 1968. The role of glucose in stimulating germination of Fomes igniarius var. populinus basidiospores. Phytopath. 59: 293-296.

53. Marsh, P.B., Taylor, E.E. and L.M. Bassler. 1959. A guide to the literature on certain effects of light on fungi: reproduction, morphology, pigmentation, and phototropic phenomena. Pl. Dis. Rep. Suppl. #261 251-312.

54. McCracken, F.I. 1974. Effects of some factors on Pleurotus ostreatus spore germination. Proc. Am. Phyto. Path. Soc. 1: 63.

55. Merrill, W. 1970. Spore germination and host penetration by heartrotting Hymenomycetes. Ann. Rev. Phytopath. 8: 281-300.

56. Merrill, W. and D.W. French. 1966. Colonization of wood by soil fungi. Phytopath. 56: 301-303.

57. Miller, D.J. 1979. Deterioration of logs in cold decks: A survey of the information applying to the Pacific Northwest. For. Prod. J. 29(1): 34-40.

58. Morton, H.L. 1964. The establishment of wood-rotting fungi by spores and mycelium. MS thesis. Univ. of Minn.

59. Morton, H.L. and D.W. French. 1966. Factors affecting germination of spores of wood-rotting fungi on wood. For. Prod. Jour. 16(3): 25-30.

60. Morton, H.L. and D.W. French. 1974. Stimulation of germination of Polyporous dryophilus basidiospores by CO . Phytopath. 64:153-154.

61. Nicholas, D.D. editor. 1973. Wood deterioration and its prevention by preservative treatments. Vol. I. Degradation and protection of wood. Vol. II. Preservatives and preservatives systems. Syracuse Univ. Press, Syracuse, NY.

62. Nobles, M.K. 1943. A contribution towards a clarification of the Trametes serialis complex. Can. J. Res. 21(C): 211-234.

63. Nobles, M.K. 1965. Identification of cultures of wood inhabiting hymenomycetes. Can. J. of Bot. 43: 1097-1139.

64. Palmer, J.G. 1974. Effects of light on development of wood-rotting and other homobasidiomycetous fungi. Center for Forest Mycology. For. Prod. Laboratory. Madison, Wisconsin. 53705.

65. Panek, E. 1963. Pretreatments for the protection of southern yellow pine poles during air seasoning. Proc. Amer. Wood Pres. Assn. 59: 189-202.

66. Panshin, A.J. and C. deZeeuw. 1970. Textbook of wood technology. Vol. I. Structure, identification, uses and properties of the commercial woods of the US and Canada. McGraw-Hill Book Co. New York, NY.

67. Rayner, A.D.M. 1977. Fungal colonization of hardwood stumps from natural sources. I. Non-basidiomycetes. Trans. Brit. Myc. Soc. 69: 291-302.

68. Rayner, A.D.M. 1977. Fungal colonization of hardwood stumps from natural sources. II. Basidiomycetes. Trans. Brit. Myc. Soc. 69: 303-312.

69. Rayner, A.D.M. and N.K. Todd. 1979. Population and community structure and dynamics of fungi in decaying wood. Adv. Bot. Res. 7: 333-420.

70. Rayner, A.D.M. and M.J. Hedges. 1982. Observation on the specificity and ecological role of basidiomycetes colonizing dead elm wood. Trans. Brit. Myc. Soc. 78(2): 370-373.

71. Reynolds, G. and G.W. Wallis. 1966. Seasonal variation in spore deposition of Fomes annosus in coastal forest of British Columbia. Can. Dep. Fish. and Forest. Bi-Mon. Res. Notes. 22(4): 6-7.

72. Reynolds, G. and H.M. Craig. 1968. Seasonal variation in infection of Douglas-fir logs and stumps by Fomes annosus. Can. Dep. Fish. Forest. Bi-Mon. Res. Notes. 24(6): 49-50.

73. Richards, C.A. and M.S. Chidester. 1940. The effects of Peniophora gigantea and Schizophyllum commune on the strength of southern yellow pine. Proc. Amer. Wood Pres. Assn. 36:24-31.

74. Rishbeth, J. 1958. Detection of viable air-borne spores in air. Nature. 181: 1549.

75. Robbins, W.J. and A. Hervey. 1960. Light and the development of Poria ambigua. Mycologia. 52: 231-247.

76. Scheffer, T.C. 1964. Biological observations of significance for improved preservative treatment. Holzforschung. 18(3): 88-94.

77. Scheffer, T.C. and R.M. Lindgren. 1936. The effect of steaming on the durability of unseasoned sap-gum lumber. Jour. For. 34: 147-153.

78. Scheffer, T.C. and W.E. Eslyn. 1961. Effect of heat on the decay resistance of wood. For. Prod. Jour. 11: 485-490.

79. Schein, R.D. 1964. Comments on the moisture requirements of fungus germination. Phytopath. 54: 1427.

80. Scheld, H.W. and J.J. Perry. 1970. Basidiospore germination in the wood-destroying fungus Lenzites saepiaria. J. Gen. Microbiol. 60: 9-21.

81. Schmidt, E.L. and D.W. French. 1978. In vitro sporulation of selected wood decay fungi. Internl. Res. Group on Wood Preservation. Working Group I. Document No. IRG/WP/190. IRG Secretariat. Princes Risborough Laboratory. Princes Risborough. Aylesbury. Buckinghamshire HP17 9PX. England.

82. Schmidt, E.L. and D.W. French. 1979. Sterilization method effects on germination of wood decay fungus spores observed by contact agar block method. Phytopath. 69(7): 688-689.

83. Schmidt, E.L. and D.W. French. 1979. Spore germination of Gloeophyllum trabeum on wood in relation to mass of the sample. Pl. Dis. Rep. 63(1): 30-31.

84. Schmidt, E.L. and D.W. French. 1979. CCA and sodium penta chlorophenate inhibition of basidiospore germination of decay fungi: contact agar method. Forest. Prod. Jour. 29(5): 53-54.

85. Schmidt, E.L. and D.W. French. 1983. Variation in germination percentage of basidiospores collected successively from wood decay fungi in culture. Can. J. Bot. 61(1): 171-173.

85. Schmitz, H. 1919. Studies in the physiology of the fungi. VI. The relationship of bacteria to cellulose fermentation induced by fungi, with special reference to the decay of wood. Ann. Mo. Bot. Gard. 6: 93-136.

87. Siepman, R. 1972. Fruit body formation of wood-destroying hymenomycetes in pure culture. Zeitschrift fur Pilzkunde. 36(1/2): 7-17.

88. Slysh, A.R. 1960. The genus Peniophora in N.Y. State and adjacent regions. State Uni. N.Y. Coll. Forestry Tech. Publ. 83: 1-95

89. Smith, R.B., Craig, H.M. and D. Chu. 1970. Fungal deterioration of second growth Douglas fir logs in coastal British Columbia. Can. J. Bot. 48: 1541-1551.

90. Stalpers, J.A. 1978. Identification of wood-inhabiting aphyllophorales in pure culture. Centraalbureau Voor Schimmelcultures, Baarn. Studies in Mycology 16: 1-248.

91. States, J.S. 1975. Normal basidiocarp development of Gloeophyllum saepiarium in culture. Mycologia. 67: 1166-1175.

92. Stenton, H. 1953. The soil fungi of Wicken Fen. Trans. Brit. Myc. Soc. 36: 304-314.

93. Tamblyn, N. and E.W.B. DaCosta. 1958. A simple technique for producing fruit bodies of wood-destroying basidiomycetes. Nature. (London) 181: 578-579.

94. Taylor, J.A. 1980. Pretreatment decay in poles. Proc. Amer. Wood Pres. Assn. 76: 227-255.

95. Teixeira, A.R. 1960. Characteristics of the generative hyphae of Polypores of North America, with special reference to the presence or absence of clamp connections. Mycologia. 52: 30-39.

96. Toole. E.R. 1971. Germination of spores of wood decay fungi on wood. Phytopath. 61: 88-90.

97. Tsuneda, I. 1978. Spore germination of Fomes species. M.S. thesis. Dept. of Botany. Univ. of Alberta. Edmonton, Alberta. Canada.

98. Wazny, J. 1979. Fungi used in standard test of toxicity of wood preservatives in various European countries. Interntl. Biodeter. Bull. 14(2): 51-56.

99. Whitney, R.D. 1966. Germination and inoculation tests with basidiospores of Polyporous tomentosus. Can. Jour. Bot. 44: 1333-1343.

100. Wilcox, W.W. 1978. Review of the literature on the effects of early stages of decay on wood strength. Wood and Fiber. 9(4): 252-257.

101. Williams, E.N.D., N.K. Todd and A.D.M. Rayner. 1984. Characterization of the spore rain of Coriolus versicolor and its ecological significance. Trans. Brit. Mycol. Soc. 82(2): 323-326.

102. Winston, P.W. and D.H. Bates. 1960. Saturated solutions for the control of humidity in biological research. Ecology. 41: 232-237.

103. Wright, K.H. and G.M. Harvey. 1967. The deterioration of beetle-killed Douglas-fir in western Oregon and Washington. US Forest Serv. Res. Pap. PNW 50.

104. Zabel, R.A., F.F. Lombard and A.M. Kenderes. 1980. Fungi associated with decay in treated Douglas fir transmission poles in the Northeastern U.S. For. Prod. Jour. 3 (4): 51-56.

105. Zabel, R.A., Wang, C.J.K. and F.C. Terracina. 1982. The fungal associates, detection, and fumigant control of decay in treated southern pine poles. Final report. Electric Power Research Institute. EL-2768 RP1471-1. Palto Alto, CA.

106. Zeller, S.M. 1920. Humidity in relation to moisture imbibition by wood and to spore germination on wood. Ann. Mo. Bot. Gard. 7: 51-75.

APPENDIX

APPENDIX

STATISTICAL ANALYSIS

Unless stated otherwise, tests were analyzed at the 95% confidence level (p=0.05). Significant differences are marked with a "**".

Table 1 (pg 28)

Analysis of variance (Anova) table

Source of variation	df	MS	Calculated F
between groups	6	10.48	1.26
within groups	14	1.38	

Table 4 (pg 36)

t-test

	Mean	s	n	Calculated t
Chlamydospores	70.6	25.3	9	2.32 **
	90.4	3.3	9	
Basidiospores	91.6	3.6	9	0.73
	90.1	5.1	9	

Table 5 (pg 38)

t-test

	Mean	s	n	Calculated t
Basidiospores	94.7	2.5	6	14.08 **
	33.5	10.4	6	

Anova table

Chlamydospores

Source of variation	df	MS	Calculated F
between groups	3	15033.1	61.86 **
within groups	42	243.0	

Least significant differences (LSD)

Comparison	difference between means	LSD
5 & 35	19.4	13.6
35 & 30	48.6 **	13.8
30 & 22	4.5	12.8

Table 6 (pg 44)

A chamber was dropped randomly from seven of the eight isolates to compensate for a chamber lost to contamination.

Anova table

Source of variation	df	MS	Calculated F
group	1	1993.8	1.93
isolate	6	1032.0	33.57 **
chamber	40	30.74	
error	48	54.87	

Table 17 (pg 72)

Anova table

Source of variation	df	MS	Calculated F
between groups	4	7.5 E-4	1.24
within groups	15	6.07 E-4	

Table 18 (pg 81)

t-test

	Mean	s	n	Calculated t
Power-drilled	46.7	10.2	3	0.95
Hand-drilled	56.7	15.0	3	

Table 19 (pg 84)

Anova table

Source of variation	df	MS	Calculated F
between groups	6	669.0	125.2 **
within groups	1533	5.3	

Least significant differences (LSD)

Comparison	difference between means	LSD
Unpeeled & Fresh	0.2	0.42
Fresh & 0-6	0.7 **	0.38
0-6 & 7-12	1.0 **	0.39
7-12 & 25+	1.6 **	0.43
25+ & 19-24	0.7 **	0.49
19-24 & 13-18	0.1	0.51

Table 20 (pg 86)

Anova table

Source of variation	df	MS	Calculated F
between groups	6	6092.7	15.2
within groups	54	401.6	

Least significant differences (LSD)

Comparison	difference between means	LSD
Unpeeled & Fresh	7.5	21.3
Fresh & 0-6	17.5	20.9
0-6 & 7-12	16.2	18.6
7-12 & 13-18	20.8 **	19.6
13-18 & 25+	0.2	19.2
25+ & 19-14	1.0	18.5

Figure 4 (pg 96)

Number of cores with basidiomycetes

Location on the pole

		Distance from top			Distance from butt			
Position	Top end	2 ft	4 ft	Mid	4 ft	2 ft	Butt end	total
Outer	302	311	279	279	254	235	251	1911
Inner	246	210	178	162	124	130	253	1303
total cores taken	3034	3037	2949	3043	3078	3083	2980	21204

Percent cores with basidiomycetes

Location on the pole

		Distance from top			Distance from butt			
Position	Top end	2 ft	4 ft	Mid	4 ft	2 ft	Butt end	LSD
Outer	9.9	10.2	9.4	9.3	8.2	7.6	8.4	1.8
Inner	8.1	6.9	6.0	5.4	4.0	4.2	8.4	1.5